EXHIBIT B

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Ecological Conditions in the Illinois River Watershed

**Expert Report of
Thomas C. Ginn, Ph.D.**

*State of Oklahoma et al. v.
Tyson, et al.*

Exponent®

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State of Oklahoma, et al. v. Tyson, et al.
Civil Action Number: 05-CV-0329-GKF-SAJ

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Thomas C. Ginn, Ph.D.

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Acronyms and Abbreviations

BMI	benthic macroinvertebrates
BUMP	Beneficial Use Monitoring Program
CDM	Camp, Dresser & McKee, Inc.
CERCLA	Comprehensive Environmental Response, Compensation and Liability Act of 1980
DELT	deformities, eroded fins, lesions, and tumor
DEM	digital elevation model
DOI	U.S. Department of the Interior
EPA	U.S. Environmental Protection Agency
EPT	Ephemeroptera-Plecoptera-Trichoptera
ERA	ecological risk assessment
GIS	geographic information system
HBI	Hilsenhoff Biotic Index
HSI	habitat suitability index
IBI	index of biotic integrity
LMBV	largemouth bass virus
NBI	nutrient biotic index
NBI-P	phosphorus-specific nutrient biotic indices
NRD	natural resource damages
NRDA	natural resource damage assessment
ODWC	Oklahoma Department of Wildlife Conservation
OWRB	Oklahoma Water Resources Board
TSI	trophic state index
WWTP	wastewater treatment plant

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1   Introduction

This report presents my opinions concerning the condition of biological resources in the Illinois River, its tributaries, and Tenkiller Ferry Lake in Oklahoma. Specifically, I have evaluated the available data on benthic macroinvertebrates (BMI) and fishes in the Illinois River and its tributaries above Tenkiller Ferry Lake. I have also evaluated the conditions of these resources in Tenkiller Ferry Lake.

In developing these opinions, I have been asked by counsel to address the following areas:

1. Evaluate the available information on biological conditions in the aquatic environments of the Illinois River Watershed (IRW), including the Illinois River and its tributary streams, and Tenkiller Ferry Lake.

2. Determine whether the methods described in the U.S. Department of Interior Rule for conducting natural resource damage assessments (NRDAs; 43 CFR §11) were followed by the State of Oklahoma in this matter.

3. Evaluate whether there are relationships between the density of poultry houses in the IRW and the structure of fish communities at downstream sampling sites. Evaluate the status of fish communities downstream of Cargill contract grower and breeder operations in the IRW.

4. Evaluate the approaches, methods, and conclusions reached in the report of Dr. Jan Stevenson (Stevenson 2008)

5. Evaluate the approaches, methods, and conclusions reached by Dr. Eugene Welch in the report of Cooke and Welch (2008).

2   Summary of Opinions

This section presents a summary of my opinions in this case, which are discussed in detail in the following sections of my report. These opinions are based on my observations made during visits to the IRW, my review of available information about the ecosystem, and my evaluation of information presented in the reports of Stevenson (2008), Cooke and Welch (2008), and other Plaintiffs' experts in this case.

2.1   Illinois River and Tributaries

1. The available BMI data show that healthy, diverse benthic macroinvertebrate communities exist throughout the Illinois River and its tributaries and that these communities are comparable to those at reference sites specified by the State of Oklahoma (the State). The few differences observed among sampling areas were associated primarily with proportion of urban land use.

2. The analysis of BMI communities in the Illinois River and its tributaries contained in Stevenson (2008) neglected to consider key variables that are known to affect BMI communities, including habitat quality of the stream environment and seasonal variability of the benthic community. It was apparent that there were significant seasonal and habitat-related effects on the BMI community structure between 2006 and 2007; these were ignored by Dr. Stevenson and instead incorrectly attributed to nutrient enrichment.

3. Based on Dr. Stevenson's own analysis of the benthic invertebrate data, there was no relationship between the assumed density of poultry operations and the multiple BMI measures he evaluated. Relationships between total phosphorus and BMI metrics reported in Stevenson (2008) were based on flawed statistical analysis using only a subset of the available data. Dr. Stevenson was unable to statistically connect the other indicators of nutrient enrichment he used (e.g., algal metrics, pH, and dissolved oxygen) to poultry house density; consequently, relationships identified between these secondary indicators and BMI metrics cannot be attributed to poultry house effects.

4. Based on the available fish data collected by the Plaintiffs, a healthy fish community exists within the IRW streams, which includes a wide diversity of game fish species. There is also an ample food base for these game fish, including a diverse population of forage fishes.

5. The State uses what is called the fish index of biotic integrity (IBI) to gauge whether a stream will support a cool water aquatic community. The IBI score encompasses a wide range of measures to predict the health of a fish community. In contrast, the Plaintiffs' expert, Dr. Stevenson, ignored this fish index in his analysis. Most of the IBI scores within the IRW streams (83–100 percent depending upon the year and data set) were at or above a score of 37, indicative of a fully supported cool water aquatic community, or had scores between 30 and 36, where a conclusive decision regarding attainment of a cool water aquatic community requires further investigation. The remaining IBI scores indicated that there were stations (0–17 percent depending upon the year and data set) that would not be predicted to support a cool water aquatic community because the IBI scores were 29 or lower.

6. Based on the Plaintiffs' Beneficial Use Monitoring Program (BUMP), the majority of the IRW streams fully supports the Fish and Wildlife Propagation beneficial use. The Fish and Wildlife Propagation beneficial use means that a stream supports a cool water aquatic community including fish and other aquatic organisms (e.g., benthic macroinvertebrates) and wildlife. Dr. Stevenson did not consider the Plaintiffs' BUMP data in his report. The State monitors six streams within the IRW as part of the BUMP to evaluate whether or not streams are attaining the Fish and Wildlife Propagation beneficial use. Based on the BUMP stream reports from 2001 to 2007, five of the six locations monitored (83 percent) attained the Fish and Wildlife Propagation beneficial use status consistently over the past 5 to 6 years.

7. Based on a U.S. Environmental Protection Agency (EPA) study of streams in the Arkansas portion of the IRW, the fish community appears unimpaired except in areas where urban influences and habitat alteration may be affecting the streams. Seven of ten stream sampling locations in the Arkansas portion of the IRW were unimpaired with regard to the fish community, including two locations along the Illinois River near the border with Oklahoma. The remaining three locations were located near urban areas of Osage Creek

and two of these three locations had impaired habitat quality compared to the reference stations.

8. Based on the 2007 fish data collected by the Plaintiffs, there is no connection between upstream poultry house density and any of the indicators of fish community health (e.g., species richness, fish IBI scores). Relationships between poultry house density and fish metrics presented in Stevenson (2008) were based on flawed statistical analysis using only a subset of the available data. Many fish metrics, including the fish IBI scores, were related to a stream's sub-basin size (i.e., size of the stream), showing the importance of the size of the sub-basin in evaluating the characteristics of fish communities.

9. Dr. Stevenson conducted an incomplete and scientifically flawed assessment of fishes in streams of the IRW. His conclusion alleging 20 percent negative effect on fish communities caused by poultry house density is based on a flawed compilation of multiple statistical relationships. Stevenson bases his 20 percent estimate on statistical models for fish metrics that primarily have no statistically significant relationship to poultry house density. In fact much greater changes (i.e., as great as 83 percent change) in the fish metrics are statistically related to sub-basin size within the IRW. Thus, the alleged 20 percent change in fish metrics is meaningless and well within limits of change resulting from a natural factor such as the size of the sub-basin within the IRW.

2.2   Tenkiller Ferry Lake

10. The simplistic evaluation of habitat "squeeze," as conducted by Cooke and Welch (2008), does not provide a scientifically valid demonstration that fish populations are being injured by eutrophic conditions in Tenkiller Ferry Lake. Specifically, Dr. Welch's opinions are based on a flawed assessment of baseline using an inappropriate reference lake (Broken Bow Reservoir) and an invalid assessment of causation concerning alleged releases of phosphorus from poultry operations.

11. For the limited number of fish species considered in his assessment, Dr. Welch fails to evaluate important habitat factors that are known to limit populations of those species other than dissolved oxygen and temperature. Cooke and Welch (2008) also fail to consider

important differences in those habitat factors between Tenkiller Ferry Lake and Broken Bow Reservoir.

12. Given the severe limitations of the assessment by Cooke and Welch (2008), there is no valid scientific basis for concluding that poultry litter application in the IRW is currently endangering fish and aquatic life in Tenkiller Ferry Lake, as is alleged by Dr. Welch.

13. Contrary to Welch's (2008) opinions, the available data indicate that Tenkiller Ferry Lake supports abundant and healthy fish populations that are characteristic of the lake's trophic status. The available information does not indicate that fish populations in Tenkiller Ferry Lake have been injured by phosphorus loading to the reservoir, regardless of the source of phosphorus.

2.3   Overall Assessment

14. The part of the complaint for this case that describes phosphorus and phosphorus compounds as hazardous substances under the Comprehensive Environmental Response, Compensation and Liability Act of 1980 (CERCLA) is misleading and inaccurate from a scientific and toxicological perspective. Phosphorus in its elemental form (P), sometimes referred to as "white phosphorus," is highly reactive and does not occur naturally in the environment. It is a highly toxic substance. Alternatively, phosphorus compounds that exist in the natural environment are very different substances. Common forms of phosphorus compounds include organic phosphorus compounds that occur in living organisms and phosphates (PO_4) that are essential nutrients for plants and animals. As an ecotoxicologist, I do not consider phosphorus as it exists in compound form in the natural environment (e.g., ortho phosphate) to be a hazardous substance as the term is used in CERCLA. Therefore, no injury as defined as part of an NRDA under CERCLA can result from exposure to naturally-occurring phosphorus compounds in the aquatic or terrestrial environments. Notwithstanding this opinion, I am proceeding to evaluate the Plaintiffs' experts claims that injuries result from phosphorus releases in the IRW and I also present my opinions concerning any adverse effects of phosphorus on fishes and invertebrates.

15. A fundamental flaw in the assessments by both Stevenson (2008) and Cooke and Welch (2008) is the lack of valid reference sites for comparison with the IRW streams and Tenkiller Ferry Lake. In both of these studies, the reference areas selected in lakes and streams are not demonstrated by the Plaintiffs' experts to be comparable to the assessment areas within the IRW for all factors except the potential influences of poultry litter application. Therefore, any conclusions reached by the authors are invalid with regard to injury resulting from poultry litter, and do not represent a scientifically defensible comparison for determining causal relationships with potential effects of poultry litter applications.

16. The assessment of biological conditions conducted by the Plaintiffs does not constitute an NRDA as described in the U.S. Department of the Interior (DOI) rule. In the case of fishes and BMI, the Plaintiffs' experts:

    - Did not provide a scientifically-valid description of baseline conditions

    - Did not use appropriate reference areas for comparison with the IRW streams and Tenkiller Ferry Lake

    - Did not conduct a valid determination that an injury exists in IRW streams or Tenkiller Ferry Lake that is caused by the release of nutrients from poultry litter to the aquatic environment

    - Did not quantify any reductions in natural resource services for the IRW streams and Tenkiller Ferry Lake that are caused by any releases of nutrients from poultry litter.

17. In addition to not complying with the requirements for an NRDA as described in the DOI Rule, the Plaintiffs' experts, Drs. Stevenson and Welch, did not conduct a reliable, systematic, and valid determination of whether any releases of phosphorus from poultry litter application had caused adverse effects on invertebrates and fishes in the Illinois River, tributary streams, and Tenkiller Ferry Lake. The DOI rule provides a reasonable framework to assess injuries in the context of an NRDA. However, if the DOI rule is not followed in an assessment, scientists should nonetheless follow certain standards of scientific practice such

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as reference area comparisons, valid statistical approaches, and assessments of causation. Drs. Stevenson and Welch failed to conduct valid scientific assessments of the status of invertebrate and fish communities in the IRW and failed to develop any reliable causal links with any phosphorus inputs from poultry operations.

18. Taken as a whole, the biological data for the Illinois River and its tributaries show that the aquatic environment supports diverse and healthy assemblages of invertebrates and fishes. The fish community of Tenkiller Ferry Lake is abundant, diverse, and supports significant sport fisheries. These biological communities do not show signs of injury that is attributable to nutrient inputs.

3   Qualifications

I am a Principal Scientist in the EcoSciences practice at Exponent, a scientific and engineering consulting firm headquartered in Menlo Park, California. I am associated with Exponent's Phoenix, Arizona, office. I have held the position of Principal Scientist at Exponent since 1997. From 1987 to 1997, I held the positions of Vice President and Principal at PTI Environmental Services, which was acquired by Exponent. As a Principal of the firm, I provide program management and expert consulting services, with primary expertise in the areas of ecological risk assessment (ERA) and NRDA.

My education is in the fields of biology and fisheries. I received a Ph.D. in biology, with a specialty in estuarine ecology, from New York University in 1977, an M.S. in biological sciences (specializing in marine biology) from Oregon State University in 1971, and a B.S. in fisheries science from Oregon State University in 1968.

I am a member of the American Chemical Society, the Society of Environmental Toxicology and Chemistry, and the American Institute of Fishery Research Biologists. I am a Certified Fisheries Professional by the American Fisheries Society, Certificate No. 2844.

My consulting experience has focused on the effects of hazardous substances on aquatic and terrestrial organisms. I have conducted studies of the effects of inorganic and organic chemicals on biological communities at many sites nationwide. I have specialized expertise in assessing the fate, exposure, and effects of substances such as arsenic, cadmium, chromium, copper, lead, mercury, zinc, polychlorinated biphenyls, polycyclic aromatic hydrocarbons (PAHs), and dioxins/furans. I have directed investigations of the biological effects of chemicals in aquatic sediments at many sites. These investigations have included the design of sampling studies and the scientific interpretation of study results.

I have also conducted studies of the effects of lake eutrophication at several sites. For EPA, I conducted an evaluation of water quality and biological conditions and potential lake restoration activities at Lafayette reservoir in California. I also conducted studies of hypereutrophic

conditions and restoration alternatives at Upper Klamath Lake in Oregon. I have also conducted ecological investigations in the following river systems: Hudson River (New York), St. Lawrence River (New York), Ashtabula River (Ohio), Ottawa River (Ohio), Clark Fork River (Montana), Coeur d'Alene River (Idaho), and the Tittabawassee and Saginaw rivers (Michigan).

I have served on scientific advisory committees for several federal government programs concerning issues of biological effects of chemicals in sediments. The dates and committees are as follows:

- 1988–1991. Member of the Technical Advisory Committee for EPA's Puget Sound Estuary Program

- 1993–1995. Member of the Technical Advisory Group for the Long Term Management Strategy, a multi-agency program for San Francisco Bay

- 1994–1996. Member of the Benthic Resource Assessment Group, a scientific advisory committee for the U.S. Army Corps of Engineers for New York/New Jersey Harbor.

I have published many articles and book chapters on various aspects of pollution biology, including serving, since 1983, as co-author for an annual review of important studies in the area of marine pollution published annually by the Water Environment Federation.

Further information on my qualifications, publications, and prior testimony is provided in Appendix A.

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5   Ecological Conditions in the Illinois River Watershed

5.1   Overview

The IRW drains an area of approximately 1,598 mi^2 and contains an extensive series of streams. The IRW is located in the northeastern corner of Oklahoma and the northwestern corner of Arkansas (see Figure 5-1). The streams within the IRW that have been under investigation have ranged from small streams with very small sub-basins of less than 10 mi^2, which are generally unnamed streams, to large streams with sub-basins of hundreds of square miles. The Illinois River has the largest sub-basin, and is the predominant stream within the IRW. Its headwaters are located in Arkansas. The Illinois River flows to the west through Arkansas, entering Oklahoma in the central portion of the IRW. Approximately 70 miles of the Illinois River have been designated by the Oklahoma Legislature as a Scenic River Area. It then flows west through Oklahoma until it bends and flows in a more southerly direction. Near the southern boundary of the catchment, the Illinois River empties into Tenkiller Ferry Lake.

The streams under investigation within the IRW are primarily classified into the cool water aquatic community subcategory with regard to their Fish and Wildlife Propagation beneficial use, as specified in Title 785 (OAC 785:45, Appendix A). A single stream (Park Hill Branch) that was sampled was classified into the warm water aquatic community subcategory with regard to its Fish and Wildlife Propagation beneficial use. Each of these subcategories has different biological criteria that the state uses to determine if the beneficial use of the stream is fully supported or not. The biological criteria used to make this determination are described in Section 5.3.

Because of the complexity of the IRW, which ranges from very small streams to the main stem of the Illinois River, it is important to be able to categorize and describe the various sizes of the streams, especially as they relate to biological conditions. Sub-basins are used to quantify the size and complexity of individual streams within a watershed, and generally, the size of a

sub-basin increases in a downstream direction within the IRW. A sub-basin is the area of land that encompasses the potential flowing surface waters above a particular point in a stream. Headwater streams that are fed directly from snow melt, outfalls, and springs have the smallest sub-basins. Physical gradients in a watershed are an important factor for describing available habitat and the size of a sub-basin is generally related to the amount of stream flow for a given area. However, this does not mean that all precipitation falling on a sub-basin necessarily flows into the stream. For the purposes of my assessment, I have determined the size of sub-basins above particular biological sampling points on the IRW streams. Therefore, the size of a stream's sub-basin provides a quantification of potential stream habitat that may be correlated with biological parameters (Vannote et al. 1980).

Sub-basins for each biological station were determined with ESRI ArcGIS software (with Spatial Analyst Extension) using a 10 m resolution National Elevation Dataset (NED) from the U.S. Geological Survey (http://seamless.usgs.gov/index.php) (ESRI 2009). NED contained a Digital Elevation Model (DEM) and elevation data. Using the DEM and the elevation data contained in the NED, the direction of stream water flow to each of the biological stations was determined. The area encompassing the streams that flow to each biological station was delineated as the sub-basin for that particular station. Stream station coordinates used to define these sub-basins were taken from two sources: 2006 River Sampling field notebook (STOK0035060–STOK0035195) and Dr. Olsen's IllinoisMaster.mdb (Olsen00004250).

For the purposes of this assessment, streams were classified on a relative basis as being large (i.e., having a total sub-basin greater than 100 mi^2), medium (sub-basin of 20–100 mi^2), small (sub-basin of 10–20 mi^2), or very small (sub-basin less than 10 mi^2). These relative classifications are also used in this report to describe the relative size of the stream at the biological sampling stations used by CDM. A sub-basin was calculated for each sample station, which represented the sub-basin within the IRW upstream of that sample station.

5.1.1   Streams of the IRW

The **Illinois River** begins as a large stream in the western portion of Arkansas and then becomes increasingly larger as it flows northeast through Arkansas. As the Illinois River begins to flow in a westerly direction through Arkansas, it joins the Osage Creek, at which point it becomes the largest stream within the IRW for the remainder of its course until it empties into Tenkiller Ferry Lake, which is the furthest downstream water body within the IRW.

Osage Creek is a large stream located in the northwest portion of Arkansas in the upper reaches of the IRW. It joins the Illinois River in the northeastern portion of the watershed.

Baron Fork is a large stream that has its headwaters in Arkansas near the southern portion of the watershed. Over a short distance, it transitions from a relatively small stream to become a large stream. Approximately 35 miles of Baron Fork within Oklahoma have been designated by the Oklahoma Legislature as a Scenic River Area. Baron Fork flows to the west for many miles as a large stream within the watershed, until it joins the Illinois River just upstream of Tenkiller Ferry Lake.

Caney Creek is a large stream at its most downstream reach within the IRW. The headwaters of Caney Creek, where it is a much smaller stream, are located in Oklahoma in the south-central portion of the IRW. Caney Creek flows to the west and then to the southwest, where it flows into Tenkiller Ferry Lake.

Bush Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Bush Creek begin as a small stream located in Arkansas in the south central portion of the IRW. Bush Creek flows south and transitions over a short distance from a small to a medium sized stream before flowing into Baron Fork, still within Arkansas.

Ballard Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Ballard Creek are located in the Arkansas in the central portion of the IRW. Ballard Creek flows from the western boarder of Arkansas to the northwest into Oklahoma

where it flows into the Illinois River near Lake Francis, located in the north central portion of the IRW.

Bidding Creek is a medium sized stream at its most downstream reach within the IRW. Bidding Creek is located entirely within Oklahoma within the west-central portion of the IRW. It flows to the southwest until it empties into Caney Creek.

Cincinnati Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Cincinnati Creek are located in Arkansas in the central portion of the IRW. It flows to the north-northwest to where it joins the Illinois River. The entire creek is contained within Arkansas.

Evansville Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Evansville Creek are located in Arkansas in the far south-central portion of the IRW. It flows to the west and then to the north through the IRW where it flows into Baron Fork in eastern Oklahoma.

Flint Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Flint Creek are located in Arkansas in the northeastern portion of the IRW. Flint Creek flows to the west into Oklahoma, where it joins with Sager Creek and continues to flow to the west until it flows into the Illinois River in the northwestern portion of the IRW. Approximately 12 miles of Flint Creek within Oklahoma have been designated by the Oklahoma Legislature as a Scenic River Area.

Fly Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Fly Creek are located in Arkansas in the south central portion of the IRW. Fly Creek flows a short distance to the west, where it flows into Baron Fork near the western border of Arkansas.

Muddy Fork is a medium sized stream at its most downstream reach within the IRW. The headwaters of Muddy Creek are located in western Arkansas in the far eastern portion of the

IRW. Muddy Creek flows in a northerly direction and joins the Illinois River in far western Arkansas.

Peacheater Creek and **Shell Branch** are both medium sized streams at their most downstream reaches within the IRW. The headwaters of Peacheater Creek and Shell Branch are located in eastern Oklahoma in the central portion of the IRW. Each creek flows in a southwesterly direction, then joins Baron Fork in the central portion of the IRW. Peacheater Creek is located further downstream along the Baron Fork than Shell Branch.

Spring Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Spring Creek are located in western Arkansas in the far northeastern portion of the IRW. Spring Creek flows in a westerly direction a short distance, then flows into Puppy Creek in western Arkansas.

Tyner Creek is a medium sized stream at its most downstream reach within the IRW. The headwaters of Tyner Creek are located in eastern Oklahoma in the west-central portion of the IRW. Tyner Creek flows in a southwesterly direction a short distance, then flows into Baron Fork well upstream of the Illinois River.

Tahlequah Creek is a small stream at its most downstream reach within the IRW and located entirely within Oklahoma. The headwaters of Tahlequah Creek are located near the far western edge of the IRW north of Tenkiller Ferry Lake. Tahlequah Creek flows in a southeasterly direction to where it empties into the Illinois River just upstream of Tenkiller Ferry Lake.

Peavine Creek is a small stream at its most downstream reach within the IRW and located entirely within Oklahoma. The headwaters of Peavine Creek are located in the west-central portion of the IRW. It Creek flows in a northeasterly direction to where it flows into Baron Fork.

Park Hill Branch is a small stream at its most downstream reach within the IRW and located entirely within Oklahoma. The headwaters of Park Hill Branch are located near the far western

edge of the IRW north of Tenkiller Ferry Lake. Park Hill Branch flows in an easterly direction to where it empties into the Illinois River just upstream of Tenkiller Ferry Lake.

5.2   Benthic Macroinvertebrates

BMI are small animals that live in or on the bottom sediments in aquatic systems such as lakes and streams. Lotic BMI (i.e., living in flowing streams) include insect larvae and nymphs (e.g., dragonflies and mayflies), oligochaetes (worms), gastropods (snails), and amphipods (scuds). Community structure of BMI is a useful indicator of stream health, because of BMI ubiquity, sedentary nature, and range of tolerances to various environmental and chemical stressors (Merritt et al. 2008). The evaluation of various BMI community metrics (e.g., proportion of sensitive taxa, abundance, and diversity, among others) is an important component of the Plaintiffs' assessment criteria for the protection of surface water (OAC 785:45-5-12 (f)).

This section of my report presents an assessment of the status of BMI in the Illinois River and tributaries at the sampling stations used by Camp, Dresser & McKee, Inc. (CDM) in 2005, 2006, and 2007. As discussed in Section 6 of this report, Stevenson (2008) does not present an overall characterization of BMI in the Illinois River system. Alternatively, he conducts only a scientifically-flawed statistical analysis and reaches confusing and inconsistent conclusions concerning the presence of any adverse effects on these biological assemblages.

In this section, several comparisons are drawn between BMI metrics in the Illinois River system and the same metrics at the Plaintiffs' designated reference streams (Little Lee Creek, Dry Creek, and Spring Creek in 2005; and Little Lee Creek in 2006 and 2007). The justification for these streams being used as appropriate reference areas is not clearly presented in the expert reports, and the use of only one or two sampling sites per reference stream is unlikely to provide a suitable comparison for all of the streams sampled in the Illinois River system. According to Olsen (2008), reference sites were suggested by a committee consisting of the Oklahoma Water Resources Board (OWRB) and Oklahoma Department of Wildlife Conservation (ODWC), and were selected as the least poultry-impacted area streams. The selected sites had low total

phosphorus levels, and "topographic data were used to calculate stream gradient and stream order to ensure that reference streams were similar in size and habitat conditions as the potentially impacted streams in IRW" (Olsen 2008). It must be noted that in making these comparisons, I am in no way indicating that I agree on the appropriateness of the Plaintiffs' designated reference streams. In fact, given the few reference streams that were sampled and the lack of hydrologic diversity compared with the Illinois River system streams, it is apparent that the reference sampling conducted by CDM was inadequate.

5.2.1   Available Databases

The 2005, 2006, and 2007 BMI data were taken from two sources: from Olsen's IllinoisMaster.mdb (2005 data) and Stevenson's CDMBenthicAllwithNames2005-2007.xls (2006 and 2007 data). The latter file was compared to BMI data downloaded from the Olsen Master database and the two files were determined to contain the same benthic data counts. This information was used to calculate a number of community metrics at each sampling station, including abundance, taxa richness, Shannon Diversity index, Ephemeroptera-Plecoptera-Trichoptera (EPT) richness, abundance, and proportional abundance of sensitive taxa, per Chapter 785 of Oklahoma Administrative Code (OAC 785:45-5-12(f)(5)). Benthic abundance is a measurement of sampled habitat health and productivity, while taxa richness (number of unique taxa collected) acts as a measure of community diversity. Shannon Diversity indices combine abundance and taxa richness data to describe how the total benthic abundance is distributed among representative taxa; degraded habitat conditions generally result in benthic communities characterized by low diversity and evenness. Measurements of benthic community sensitivity (EPT taxa richness, EPT relative abundance, and biotic indices that incorporate intolerance values) are also useful, because nutrient enrichment and/or habitat degradation can reduce the relative abundance and diversity of intolerant benthic invertebrates.

Land use information and sub-basin size were generated for each station with DEM using the geographic information system (GIS). The results of these calculations are summarized in Tables 5-1 to 5-3.

5.2.2   Data Limitations

The 2005 BMI data collected by CDM were limited by the small sample sizes. Sampling was conducted at a total of 13 sites, ten of which were located within the Illinois River system. Sites were not randomly selected, but were chosen based on prior stream sediment sampling results to provide a range of potentially impacted sites (Olsen 2008; Stevenson 2008).   Although the lack of randomization and the lack of replicate samples in the study design could have implications for certain statistical analyses, the data are nonetheless sufficient to provide an overall characterization of community structure at the study sites. Benthic metrics were calculated for these data to provide a general, qualitative indicator of community health and diversity for comparison with later sampling efforts.

The major limitation of 2006 and 2007 BMI data is the lack of habitat and substrate information. This is a significant study design limitation for the CDM data. Stream habitat assessments were performed for each site during the preliminary 2005 IRW biological survey, and information recorded at the sampled stations included stream width, flow, depth, riparian habitat, substrate characteristics.

However, habitat assessments were apparently not taken during subsequent sampling events conducted during 2006 and 2007. Because the number of biological sampling stations was greatly expanded for these later events (13 total in 2005 versus 72 in 2006 and 70 in 2007), there is a lack of both recent and past habitat condition information at these additional sampling sites. Without site-specific physical habitat assessments, it is impossible to judge to what degree 2006 and 2007 benthic community variability is linked to physical variability in the habitat quality.

In conducting assessments of stream BMI, it is recognized that habitat and sediment characteristics are very important in determining the abundance and structure of these assemblages. Without this information, there will always be a great deal of uncertainty concerning the cause(s) of any apparent differences between community metrics at alternative locations. Documents describing the standards of scientific practice for BMI assessments commonly stress the importance of data on stream habitat, including sediment type, in the interpretation of the resultant data (Beisel et al. 1998; Crunkilton and Duchrow 1991; Erman

and Erman 1984; Rempel et al. 2000; Richards et al. 1993; Stark 1993; Vinson and Hawkins 1998).

5.2.2.1   Sample Types

Sampling events in 2005, 2006, and 2007 were planned and conducted by CDM. BMI communities were sampled in August and September of 2005, August 2006, and April 2007. As described below, the collection methodology varied significantly among different sampling events.

2005 Sampling Event—During the biological survey conducted in 2005, BMI communities were sampled using D-ring dip nets (also called D-nets), kick nets, and artificial substrate Hester-Dendy samplers. Three kick net collections were conducted in a riffle area of the 100-m stretches: one in an area with large rocks and little interstitial silt, one in an area with large amounts of silt and smaller rock substrate, and the third within an intermediate riffle habitat (Brown 2008, STOK0020899). One D-net sample was taken each from submerged vegetation and from submerged woody debris. Finally, three Hester-Dendy artificial substrate samplers were deployed at each station in areas with current of greater than 0.2 ft per second and sufficient depth to ensure complete submersion over the 6-week period. It is unclear from SOPs whether these were deployed prior to or during the kick net and D-net sampling events. Collected macroinvertebrate samples were preserved in 100 percent ethanol prior to shipping and identification (Brown 2008, STOK0020896).

2006–2007 Sampling Events—Because the number of sampling stations was increased five times over that of 2005, sampling methodology used in 2006 and 2007 was simplified to allow for a more rapid characterization of BMI communities. In both years, three 1-m^2 kick net samples were taken from randomly selected locations at each sampling site. Substrate within the kick net area was agitated for a set time, and collected invertebrates were preserved in isopropyl alcohol prior to shipping and laboratory identification (Brown 2008, STOK0020917).

5.2.2.2   Spatial and Temporal Coverage

A total of 96 Illinois River system BMI sampling sites were sampled over this 3-year period, with an additional five stations located outside of the watershed as reference sites. Thirteen sites were sampled in 2005, and 72 and 70 total sites were sampled in 2006 and 2007, respectively. The location of these sampling sites within the IRW for these years is depicted in Figures 5-2 through 5-4).

2005 Sampling Event—BMI samples were collected at 10 stations during the 2005 preliminary biological survey of the Illinois River system; benthic collections were also taken at three reference sites (Table 5-1). Sampling locations were targeted to encompass 10 impacted locations within the tributaries and streams of the Illinois River system (Figure 5-2) (Brown 2008, STOK0020895). Two reference sites were located immediately adjacent to the Illinois River system (Little Lee and Spring creeks), but outside of the watershed; the third was situated in Dry Creek within Arkansas' Buffalo River system. Stations BS-08, BS-208, BS-28, BS-62A, BS-68, BS-HF22, BS-HF28A, BS-HF04, BS-REF1, and BS-REF2 were sampled in August, while the remaining stations were sampled the following month, according to the Olsen database.

2006 Sampling Event—During the 2006 secondary biological screening, BMI communities were sampled at a total of 72 stations. Sixty-nine stations were located within the Illinois River system, with specific site selection based on proximity to areas with high poultry house density and the results of the 2006 Water Quality Investigation (Figure 5-3). An additional sampling station located downstream of Tenkiller Ferry Lake was not included in benthic community analyses, because this station was unlikely to be representative of biological conditions in IRW streams. Two reference sites were located in Little Lee Creek, adjacent to the Illinois River (Table 5-2). All collections were taken between August 8 and August 16, 2006.

2007 Sampling Event—In 2007, BMI community samples were collected from total of 70 stations (Table 5-3), 46 of which were also sampled in 2006. Sixty-eight stations were located in the Illinois River system, with specific site selection based on data gathered in 2006 along with proximity to impacted areas (Figure 5-4). Two reference sites were located in Little

Lee Creek. Unlike previous sampling events, 2007 benthic community collections took place in the spring, between April 4 and April 22. Because "streams often exhibit a large amount of seasonal variability in the structure and composition of benthic macroinvertebrate communities," these data are of little use for a year-over-year comparison with 2005 and 2006 benthic data (Brown 2008, STOK0020895).

5.2.2.3   Taxonomic Levels

According to the SOPs included in the Darren Brown expert report (Brown 2008) identification of collected BMI was carried out to the lowest possible taxonomic level. As a result of diverse and varied BMI morphology, the refinement level for taxonomic identifications fluctuates with taxa, and it is not always possible to identify specimens to the same taxonomic level. Often, identification of more difficult specimens will not be carried out beyond the familial or ordinal level.

Of the total taxa identified in 2005, 4.2 percent were identified to the species level, and 70.5 percent to the genus level, with the remaining specimens identified to the familial level or higher. Similarly, in 2006, 6.0 percent of the specimens collected were identified to the species level, with 68.8 percent identified to the genus level. However, the number of species identified for 2007 macroinvertebrate collections increased to 19.0 percent of all identified taxa, while genus level identification was comparable to previous years. It is unclear whether this difference resulted from an alteration of taxonomic identification techniques, increased competence of the taxonomists, or if the communities sampled in April 2007 were more easily identifiable than those collected in August 2006.

5.2.3   Summary of BMI Community Data

Although either three or eight sub-samples were collected at each station (depending on the year), available data suggest that these were combined to give one aggregate sample per sampling station. Consequently, it is impossible to make any inferences regarding within-site community variability, because sub-sample data were not made available. Without information

of within-station variability, it is also not possible to conduct statistical tests of significant differences between study areas. This is another substantive deficiency of the BMI study design used by CDM. In such assessments, replicate samples are typically collected as a matter of standard practice to determine within-station variability and to make statistical comparisons among sampling locations, including comparisons with reference areas.

Notwithstanding these study design limitations that preclude statistical comparisons, the available benthic data suggest that healthy, diverse benthic communities existed at all sites sampled in the IRW during the summers of 2005 and 2006, and that these communities were comparable to those sampled at the Plaintiffs' reference sites. BMI samples taken in April 2007 showed different community composition versus summer 2005 and 2006 collections; however, this shift was evident in reference station communities as well.

Descriptions of various BMI community metrics are provided in the following sections.

5.2.3.1   Taxa Richness

The taxa richness of a benthic invertebrate sample is simply a count of the number of unique taxa collected at each site, and as such, depends on the level of taxonomic identification and the standardization of identification methodology. Taxa richness is a reflection of sample community diversity, and has been shown to negatively correlate with increased habitat degradation and nutrient enrichment. EPT taxa richness is also an important indicator in assessing these effects on BMI, as these taxa are generally sensitive to water quality impacts, including nutrient effects (Barbour et al. 1999).

Between 15 and 38 unique macroinvertebrate taxa were collected at Illinois River system sampling sites in 2005; taxa richness at reference sites BS-REF1, BS-REF2, and BS-REF3 was 33, 24, and 26, respectively. Similarly, the numbers of unique EPT taxa (i.e., members of the orders Ephemeroptera, Plecoptera, and Trichoptera) collected at 2005 Illinois River system sites was comparable to those collected at reference sites (Figure 5-5, Table 5-4).

Extensive sampling during the summer of 2006 revealed taxa richness of benthic communities ranging from 6 to 39 unique taxa (median taxa richness = 23; average taxa richness = 23.06 ± 6.13) in Illinois River system sites. Taxa richness was 28 and 30 at the two reference stations sampled (Figure 5-6). EPT taxa richness at Illinois River system sites ranged from 0 to 15 (median = 8; average = 8.01 ± 3.19), whereas EPT taxa richness was 11 at both reference sites (Figure 5-7, Table 5-4). Commonly collected mayfly (Ephemeroptera) families included Baetidae, Caenidae, Leptophlebiidae, and Heptageniidae, largely intolerant to moderately intolerant scraper and collector taxa that inhabit lotic-erosional (cobble and gravel areas within streams and rivers) and lotic-depositional (pools and bank margins within streams and rivers) habitats. Caddisfly (Trichoptera) communities were dominated by moderately tolerant to intolerant Hydropsychidae and Philopotamidae individuals, collector-filterer taxa that occupy lotic-erosional habitats. Other collected taxa include warm river-associated Philopotamidae species, predaceous Polycentropodidae taxa, and intolerant Helicopsychidae scrapers. Stonefly (Plecoptera) individuals were more infrequently sampled than either Ephemeroptera or Trichoptera taxa at both Illinois River system and reference sites, and all families collected in 2006 (Perlidae, Capniidae and Leuctridae) are characterized as highly intolerant taxa.

Taxa richness at 68 Illinois River system sites ranged between 8 and 47 for spring 2007 benthic sampling (median = 28; average = 27.49 ± 7.6); 21 and 35 unique taxa were collected at reference sites (Figure 5-8). Between 1 and 18 unique EPT taxa were collected at Illinois River system sampling sites during spring 2007 (median = 8.5; average = 9.16 ± 4.62); 10 and 12 unique EPT taxa were collected each at the two reference sites (Figure 5-9, Table 5-4). The taxonomic make-up of 2007 EPT communities was similar to that of 2006, with the addition of the Ephemeropteran family Ephemerellidae (intolerant collector-gatherers), and the Trichopteran family Glossosomatidae (intolerant scrapers). Three additional Plecoptera taxa were collected in 2007: Chloroperlidae (intolerant predators), Nemouridae (moderately intolerant shredders), and Perlodidae (intolerant predators). These changes in EPT community composition most likely occurred as a result of the shift in seasonal sampling time from late summer to spring.

5.2.3.2   Diversity

The Shannon Diversity index combines taxa richness and the distribution of collected specimens within the taxonomic groups to give a measure of community diversity and evenness. The Shannon Index is the most commonly used metric in aquatic ecosystem analysis (Lydy et al. 2000); for aquatic communities, these indices are negatively correlated with increasing habitat degradation and nutrient enrichment (Barbour et al. 1999).

Shannon indices for Illinois River system BMI communities sampled in the summer of 2005 ranged between 1.74 and 2.97. Benthic surveys conducted in 2006 produced a range of 1.20 to 3.08 for benthic community Shannon indices (average = 2.21 ± 0.35). Sampling efforts in the spring of 2007 indicated a range of Shannon indices between 1.20 and 2.99 (average = 2.39 ± 0.43) (Table 5-4). The similarity of ranges and averages of BMI Shannon diversity indices over sampling years and seasons indicates the presence of a stable Illinois River system benthic community composition.

5.2.3.3   Abundance

Total Abundance—The most productive and healthy benthic habitats tend to support high macroinvertebrate abundances (Barbour et al. 1999). Consequently, the number of collected specimens reflects the health of the collection site, as long as sampling methodology remains constant.

During the 2005 preliminary BMI survey, abundances at Illinois River system sites averaged 392.9 (± 131.2) individuals per site. Extensive Illinois River system benthic sampling conducted in 2006 and 2007 gave average abundances of 255.9 (± 93.1) and 370.6 (± 200.8), respectively, for the same area (Table 5-4). Because available benthic databases contained only one abundance count per 2005 sampling site, it is assumed that the 8 sub-samples were combined to give one total count. This may explain why 2005 average abundance is somewhat higher than those for subsequent years, when only three sub-samples were collected.

Relative EPT and Dipteran Abundance—Relative abundance of intolerant and tolerant taxa is also an important indicator of habitat stability and suitability, as "a healthy and stable assemblage will be relatively consistent in its proportional representation, though individual abundances may vary in magnitude" (Barbour et al. 1999). Percent EPT abundance is a commonly used and reliable metric for determining habitat perturbation; as benthic habitat degradation or nutrient enrichment increases, relative abundance of EPT taxa decreases. Lydy et al. (2000) compared a number of different BMI indices prior to and following the upgrading of wastewater treatment plants (WWTPs). Of all the indices tested, the relative abundance of EPT individuals was the best indicator of water quality improvement.

For Illinois River system samples, relative abundance of EPT individuals averaged 46.57 percent (± 14.15) in 2005, 50.36 percent (± 22.97) in 2006, and 32.64 percent (± 23.15) in 2007, indicating that the majority of these sites supported high relative abundances of EPT individuals (Table 5-4).

Unlike relative EPT abundance, the proportion of dipteran (midge) individuals positively correlates with habitat degradation; as conditions become more degraded, the relative abundance of dipteran taxa within the BMI community increases (Barbour et al. 1999). In 2005, dipteran abundance at Illinois River system sites averaged 8.04 percent (± 9.02), and in 2006 Illinois River system samples consisted of 10.05 percent (± 12.28) dipteran individuals. However, in the spring of 2007, Illinois River system benthic samples contained higher proportions of dipteran individuals than in previous years: 48.07 percent (± 27.93). In effect, average abundance increased five-fold over the eight months separating 2006 and 2007 benthic surveys. This difference may have resulted from the execution of benthic sampling in April rather than late summer, as in previous years. The high 2007 dipteran abundances at reference sites RS-10003 and RS-10004 (51.3 and 69.6 percent, respectively) provide strong evidence that increases in dipteran abundance resulted from natural seasonal effects on benthic community composition.

5.2.3.4   Indicator Species

The relative sensitivity of different BMI taxa can be gauged using assigned tolerance values (such as those used to compile the Hilsenhoff Biotic Index [HBI]). A phosphorus-specific set of tolerance values has been developed using frequency distributions of benthic taxa presence or absence at various phosphorus concentrations (Smith et al. 2007). Assigned phosphorus tolerance values range from 0 (extremely phosphorus intolerant) to 10 (extremely phosphorus-tolerant).

For 2006 benthic collections, 67 of the 69 Illinois River system sites sampled were inhabited by taxa that are generally intolerant to phosphorous contamination (tolerance value of 0, 1, or 3), including *Psephenus* spp (Coleoptera: Psephenidae), *Caenis* spp (Ephemeroptera: Caenidae), *Chimarra* spp (Trichoptera: Philopotamidae), *Leucrocuta* spp (Ephemeroptera: Heptageniidae), *Hexatoma* spp (Diptera: Tipulidae), and *Acruneuria* spp (Plecoptera: Perlidae) (Smith et al. 2007). Similarly, in 2007, 67 of the 68 Illinois River system samples collected contained phosphorus-intolerant taxa, including *Orthocladius* spp (Diptera: Chironomidae), *Micropsectra* spp (Diptera: Chironomidae), Paraleptophlebia *spp* (*Ephemeroptera: Leptophlebiidae*), *Stempellinella* spp (Diptera: Chironomidae), and *Rhithrogena* spp (Ephemeroptera: Heptageniidae).

HBI values were calculated for benthic communities sampled in 2006 and 2007. Scores were not calculated for the 13 sites sampled in 2005, given the preliminary nature of the survey and variety of sampling methodologies utilized. HBI scores are calculated by weighting the abundances of various benthic taxa by their respective nutrient tolerances, resulting in numeric values ranging from 0 (highly intolerant) to 10 (highly tolerant). These weighted abundances are then summed and divided by total abundance, giving a total community HBI (Hilsenhoff 1988a). Higher community HBI scores signify the presence of a tolerant benthic community, whereas low scores are indicative of invertebrate communities dominated by taxa relatively intolerant of nutrient enrichment. Tolerance values were assigned to the lowest possible taxonomic level available for each specimen count, using Hilsenhoff (1987), in addition to Midwest tolerance values listed in Barbour et al. (1999), Bode et al. (1991), and Hilsenhoff (1988a). The average HBI score for 2006 Illinois River system benthic communities was

4.75 (± 0.57), with reference site scores of 4.15 and 4.55. Sampling in the spring of 2007 yielded Illinois River system communities with an average HBI score of 5.3 (± 1.02), and reference communities with scores of 4.54 and 4.6 (Table 5-4, Figure 5-10). According to Hilsenhoff (1987), the 2006 reference HBI scores are indicative of "very good" or "good" conditions; 64 of the 68 Illinois River system HBI scores also indicated "good" or "very good" conditions. The remaining four sites indicated "fair" conditions, which is not unexpected given the level of anthropogenic impacts within the watershed. Benthic communities sampled in 2007 exhibited greater variation in HBI scores, with an increased number indicating "fair" or "fairly poor" conditions. However, temporal differences in community structure can significantly alter HBI scores (Lydy et al. 2000; Hilsenhoff 1988b), so 2007 HBI scores may not be a reliable indicator of nutrient effects.

Phosphorus-specific nutrient biotic indices (NBI-P) were also determined for 2006 and 2007 benthic communities, using tolerance values obtained from Smith et al. (2007) and Yuan (2004). NBI-P scores are calculated by weighting the abundances of benthic taxa by their respective phosphorus tolerances. Phosphorus tolerance values range from 0 (highly intolerant) to 10 (highly tolerant). Therefore, this index increases as community composition shifts to include higher abundances of phosphorus-tolerant benthic taxa. Nutrient biotic index (NBI) values for Illinois River system benthic communities averaged 5.14 (± 0.85) and 6.48 (± 1.08), in summer 2006 and spring 2007 respectively (Table 5-4, Figure 5-11). The increase in NBI-P was most likely caused by the increase in relative dipteran abundance in 2007. A similar increase in dipteran abundance at reference station RS-10004 resulted in an NBI score increase from 3.99 in the summer of 2006 to 6.59 in the spring of 2007. Increased abundance occurred both at IRW sampling stations and at state-designated reference sites. This suggests that normal seasonal fluctuations in benthic community composition may affect biotic indices.

5.2.4   Spatial Patterns of Community Characteristics

5.2.4.1   Comparisons to Reference Areas

Sampling sites located within reference areas selected by the State are presumed to be minimally disturbed and provide a frame of reference for evaluating community metrics at potentially impacted sites. BMI sampling efforts were specifically designed to facilitate reference site versus study site comparisons: "by comparing the composition and density of macroinvertebrate populations between affected and reference streams at similar times of the year, a valuable assessment of the environmental impact of various forms of pollution can be formulated" (Brown 2008, STOK0020895; Olsen 2008, p. 2-48). However, given the temporal and methodical variability of sampling efforts in 2005, 2006, and 2007, it would be inappropriate to compare values across sampling years.

5.2.4.2   Relationship to Sub-basin Size

Aquatic community metrics, including abundance, taxa richness, and diversity, are significantly influenced by stream size and flow characteristics, especially in the case of fish communities (Dauwalter et al. 2007; Crunkilton and Duchrow 1991). Benthic macroinvertebrate communities, however, are strongly governed by microhabitat conditions (e.g., localized flow rates, benthic particle composition; [Gebler 2004]). Spearman correlations of benthic metrics with sampling site sub-basin size indicated no significant relationships between location of sampling sites within the Illinois River system and indicators of benthic community health (Table 5-5). Reference conditions were compared to those of Illinois River System sites with similar sub-basin sizes. IRW sites selected for the comparison had sub-basins ranging from 36.18 to 75.42 mi^2, the sub-basin sizes of the two reference sites. This specific evaluation should account for any natural variability that may exist between benthic communities inhabiting sites with different sub-basins, allowing for the comparison of Illinois River sites most like reference areas to conditions at those reference sites (Crunkilton and Duchrow 1991; Conquest et al. 1993).

Average abundances, Shannon diversity indices, percent EPT abundance and percent dipteran abundance at selected Illinois River system sites were similar to corresponding reference site values in all sampling years (Figures 5-12 through 5-15). It is worth noting that relative dipteran abundance significantly increased at both Illinois River system and reference sites in spring 2007; this underscores the importance of minimizing temporal variation in annual BMI surveys. Ultimately, no significant differences were detected between reference benthic communities and benthic communities inhabiting similar sites within the Illinois River system.

5.2.4.3   Comparisons to Urban Land Use

Dr. Stevenson conducted two types of correlation analyses on BMI metrics: all sampling sites and "low urban" sites. When high urban influence sites were removed from the 2007 benthic data set prior to analysis, the number of total significant correlations decreased when compared with whole data set analysis (Stevenson 2008). This is a compelling indication that urban land use may be the strongest influence on 2007 benthic community measurements. As such, eight benthic metrics (total abundance, taxa richness, EPT taxa richness, HBI, NBI-P, Shannon diversity, relative EPT abundance, and relative dipteran abundance) were regressed against percent urban land use. Statistical results are presented in Table 5-6.

Seven of the eight benthic metrics were significantly correlated with urban land use in 2007 data. HBI, NBI-P, and relative dipteran abundance all increased with greater urban influence, while taxa richness, EPT taxa richness, diversity, and relative EPT abundance were negatively correlated with increasing urban land use. The first metrics are considered positive indicators of habitat degradation, whereas the latter are negative indicators, declining with along with habitat conditions. Therefore, these sets of correlations support the previous indications that urban land use explains most of the variability in 2007 Illinois River system benthic communities.

Conversely, analysis of 2006 data identified no significant correlation between benthic metrics and urban land using Bonferroni-adjusted significance levels ($P = 0.05/8 = 0.0063$). Community NBI-P scores increased with urban land influences, but this relationship was not significant ($P = 0.037$) (Table 5-6). Other significant trends identified during the analysis of

spring 2007 data were not identified for summer 2006 benthic communities, which underscores the natural impact of season on benthic community structure. Given that BMI communities fluctuate with time of year, it is probable that the relative influence of urban land use on benthic communities varies with season as well.

5.2.4.4   BMI Communities in the Main Stem of the Illinois River

Seven stations were located within the main stem of the Illinois River during the 2006 benthic sampling event (RS-133, RS-313, RS-43, RS-657, RS-7194800, RS-7195430, RS-757). Relative abundance of EPT individuals ranged between 39.4 and 84.6 percent of total individuals collected at each station, with an average relative EPT abundance of 58.6 percent. This suggests that the benthic community of the main stem Illinois River is largely dominated by EPT individuals. In fact, average EPT abundance was higher in the Illinois River than communities in smaller tributaries and reference sites, although this was not statistically significant. Similarly, Shannon diversity indices for Illinois River benthic communities ranged from 2.15 to 2.47; communities collected from reference stations and other sampling stations within Illinois River system tributaries exhibited similar scores.

These benthic communities sampled from Illinois River sites were characterized by the following dominant taxa (Merritt et al. 2008):

1. *Caenis* spp (collector-gatherer mayflies commonly found in depositional areas)

2. *Stenonema* spp (moderately intolerant scraper mayflies inhabiting cobble areas)

3. *Tricorythodes* spp (collector-gatherer mayflies that inhabit deposition and littoral zones)

4. *Cheumatopyche* spp (filtering caddisflies commonly found in warmer rivers)

5. *Chimarra* spp (moderately intolerant filtering caddisflies commonly found in warmer rivers)

6. *Stenelmis* spp (scraper beetle larvae that inhabit coarse sediments and detritus).

These taxa were found at multiple sites and comprised a large portion of total abundances. Benthic communities collected from large riverine systems would most likely contain two or more of these taxa.

In spring of 2007, benthic community sampling involved seven total Illinois River sites (RS-133, RS-234, RS-43, RS-433, RS-654, RS-7195430, RS-757), although these were not the same seven sites sampled in 2006. Relative EPT abundances of sampled Illinois River sites ranged from 16.2 to 90.4 percent, with five out of the seven sites yielding relative EPT abundances of greater than 40 percent, and an overall average of 57.5 percent for all Illinois River sites. As a point of comparison, EPT abundances averaged 36.3 percent for reference site sampling and 29.8 percent for samples taken from Illinois River tributaries. Shannon diversity indices for Illinois River benthic samples ranged from 2.0 to 2.9, with an average index of 2.4; the lowest Illinois River diversity index, however, apparently resulted from a high relative abundance of mayflies collected at the site. Reference benthic communities and BMI communities sampled from Illinois River tributaries yielded average Shannon diversity indices of 2.2 and 2.4, respectively.

Benthic communities sampled in spring of 2007 were different in taxonomic composition than communities sampled in summer of 2006. Dominant taxa in 2007 included the following (Merritt et al 2008):

1. *Perlesta* spp (predator and early-instar collector-gatherer stoneflies)

2. *Psephenus herricki* (scraper beetle larva found in erosional lotic environments)

3. *Maccaffertium* spp (scraper mayflies that inhabit cobbled benthos)

4. *Anthopotamus verticis* (moderately intolerant burrowing, filtering mayflies)

5. Chironomidae taxa, including *Polypedilum* spp and *Cricotopus* spp;
    Chironomus spp and *Tanytarsus* spp (moderately intolerant to moderately
    tolerant shredder and collector-gatherer midge larvae that inhabit
    detritus/vascular plants).

Most likely, variability in the dominant taxa composition reflects the change in available food
sources from late summer 2006 to spring 2007 and the timing of emergences of insect larvae.

Overall, benthic metrics derived from 2006 and 2007 sampling data indicate that BMI
communities inhabiting the main stem of the Illinois River are characterized by relatively high
EPT abundances and Shannon Diversity indices, comparable to those derived from reference
sites and small Illinois River system tributaries. Benthic taxonomic composition, although
variable from 2006 to 2007, reflects expected habitat conditions for a large riverine system, with
high abundances of collector-gatherers that prefer depositional and littoral zones.

5.2.5   Summary

Overall, BMI communities sampled in the Illinois River system are indicative of a healthy,
viable ecosystem, and do not suggest evidence of degradation or stress resulting from nutrient
enrichment within the basin. Preliminary benthic sampling conducted in the summer of 2005
revealed Illinois River system communities characterized by high abundances (206 to
616 individuals collected per site), good taxa richness (15 to 38 unique taxa per site), and
excellent relative EPT abundances (up to 72 percent of total individuals collected) (Table 5-4).
Similarly, communities sampled in the summer of 2006 indicated similar characteristics, with
high BMI abundances, good taxa richness, high proportional representation by sensitive EPT
individuals, and relatively low HBI and NBI-P community scores. Benthic sampling in the
spring of 2007 revealed different benthic communities, most likely reflecting seasonal
differences. When compared with previous samples, the 2007 data displayed higher abundances
of dipteran (midge) larvae and an increase in both HBI and NBI-P scores. The effect of
seasonal dynamics on benthic community structure is well documented (Bêche et al. 2006;

Linke et al. 1999; Murphy and Giller 2000), and concurrent sampling within the reference sites in Little Lee Creek indicates a similar shift in community structure. Relative dipteran abundance at reference sites RS-10003 and RS-10004 increased from 14.2 and 25.7 percent, respectively, to 51.3 and 69.6 percent over the eight months separating 2006 and 2007 sampling events. Likewise, the NBI-P score for the BMI communities sampled at RS-10004 increased from 3.99 in August 2006 to 6.59 in April 2007. These identical and simultaneous shifts in reference community and Illinois River system community conditions strongly suggest that normal seasonal benthic dynamics are driving the changes in the individual metrics.

In addition to seasonal effects, urban land use explains much of the evident variability in 2007 benthic data. HBI, NBI-P, relative dipteran abundance, EPT taxa richness, diversity, and relative EPT abundance were all significantly correlated with percent urban land use. As increased urbanization often leads to degraded stream habitat, benthic community metrics are expected to be altered by an increase of urban land use. Overall, however, Illinois River system BMI metrics did not reflect the depauperate, low diversity benthic communities characteristic of degraded waters.

Ultimately, significant flaws in sampling methodology and implementation prevent detailed statistical evaluation of the BMI communities that were sampled by CDM. These study design deficiencies, including the lack of replicate samples and absence of key habitat data (e.g., sediment characteristics) represent significant problems with the Plaintiffs' study. Notwithstanding these limitations, a semiquantitative and qualitative analysis of the data is possible, and this assessment indicates that the Illinois River system supports an abundant and healthy BMI community.

5.3   Fishes

This section describes the fish communities inhabiting the Illinois River and its tributaries. The objective of this assessment is to evaluate the abundance and diversity of fish communities and to assess whether the fishes of these lotic environments (flowing waters) appear to be adversely affected by water quality conditions in the watershed. To accomplish this objective, I will

evaluate the structure of these communities and I will determine the relationships of various fish community metrics with watershed characteristics, including the relative density of poultry houses in upstream sub-basins. This description of the community characteristics is especially important because the report of Dr. Jan Stevenson (Stevenson 2008) does not contain such an overall assessment of the fish communities, but focuses on a complex series of flawed correlation analyses that do not provide a reliable assessment of the current status of fishes in the Illinois River and its tributaries (see Section 6 of this report).

5.3.1   Available Databases

The following are the available fish data sets that were used to evaluate the condition of the fish community within the Illinois River and its tributaries:

- 2005 fish data collected by CDM as discussed in Darren L. Brown's expert report (Brown 2008) and reported in Dr. Olsen's database (April 19, 2008).

- 2007 fish data collected by CDM as discussed in Brown (2008) and reported in Stevenson (2008) for 37 fish sample stations collected in the summer of 2007. The data used were obtained from computer files of Dr. Jan Stevenson because the data were not available in Dr. Olsen's database and only a portion of the fish field forms were available from the Plaintiffs.

- Oklahoma Beneficial Use Monitoring Reports 2001 through 2007.

- EPA Region 6 Study, *Final Report—Volume 1, Water Quality and Biological Assessment of Selected Segments in the Illinois River Basin and Kings River Basin, Arkansas* (U.S. EPA 2004).

5.3.1.1   Data Limitations

The fish data collected by CDM in 2005 were limited by the small sample size (n=10 within and n=3 outside the Illinois River system) and for this reason a rigorous statistical analysis of these data was not possible. As mentioned in Stevenson (2008), these samples were not selected

randomly, and based on that limitation alone he did not evaluate the 2005 fish data as part of his expert report. There were also no replicate samples of fish collected and only limited descriptions of the stream habitat was provided. Notwithstanding this potential limitation, I have developed fish metrics for the 2005 fish data to use for comparative purposes to the 2007 data set. The 2005 fish data provide a general indication of the health of the fish community at the specific locations sampled within the Illinois River system, but no detailed statistical analysis of the data was possible because of the limited sample size. Moreover, sampling sites were located predominantly on streams with medium sized sub-basins, excluding small headwater streams and larger streams within the IRW. For example, there was not a single fish sample collected in 2005 on the main stem of the Illinois River. For these reasons the 2005 fish data cannot be used to adequately reflect the conditions in the Illinois River system as a whole.

The sample size limitation with the 2005 fish data does not exist with the 2007 fish data set. In 2007 a total of 35 stations were sampled within the Illinois River and its tributaries over a range of sub-basin sizes (i.e., from very small to large sib-basins). These 35 stations were also reportedly selected in a random fashion and stratified over four quartiles ranging from low to high poultry house density present in the IRW. For this reason, a statistical analysis of the 2007 fish data was performed to evaluate whether or not there were statistically significant relationships between specific fish metrics and dependent variables such as size of sub-basin and poultry house density (discussed further in Section 5.3.3).

The BUMP stream reports were used as an additional point of comparison to gauge the condition of the fish community within the Illinois River and its tributaries. The BUMP reports provided a measure of beneficial use for Fish and Wildlife Propagation that can only be attained if the fish community at a station is healthy. Therefore the data from the BUMP reports were used qualitatively as discussed in Section 5.3.3. to assess the condition of the fish community in the Illinois River system from 2001–2007 (excluding 2006, for which a report was not available).

The U.S. EPA (2004) study was carefully planned, and evaluated multiple lines of evidence including water chemistry, habitat conditions, and biological conditions that included

periphyton, filamentous algae, BMI, and fish community composition. They also performed the evaluation over a number of sampling events. The primary limitation is that the evaluations of these parameters were not consistently completed at each sampling station, and so there were data gaps for some stations. Also, these data were only collected in the Arkansas portion of the IRW.

5.3.1.2   Sample Types by Year

2005 Fish Data—In 2005 fish samples were collected from ten stations within the IRW, and three additional reference sites located outside the IRW (refer to SOP 7-1 in Brown 2008).

As discussed in Olsen (2008) the reference stations used from 2005 (as well as 2006 and 2007) were selected based on a number of factors including poultry house density as follows:

> The selection of the reference biological sampling locations for 2005 was based upon a step-wise approach similar to the approach used to select the 2005 river and biological sampling stations within the IRW. The first step entailed meeting with Oklahoma Water Resource Board (OWRB) and the Oklahoma Department of Wildlife and Conservation (ODWC) to obtain their recommendations on potential reference waterbodies for streams in the IRW. Their recommendations included Spring Creek, a tributary to the Neosho River and Little Lee Creek, a tributary to Lee Creek. Both of these waterbodies are just outside of the IRW and within the Ozark Highlands ecoregion. In addition to these waterbodies, CDM sampled several other streams within the Ozark Highlands ecoregion in Arkansas. As was outlined in Section 2.2.7 of this report, data were collected from these waterbodies to determine if they were being impacted by poultry operations. Specific data evaluated include water quality and sediment analysis, soils, bedrock geology, land use data, topography, hydrologic data, groundwater data, and official findings on water quality.
>
> The density of poultry operations in sub-watersheds was an important consideration in selecting reference locations. Ideally, reference areas would have little or no poultry houses in the watershed and total phosphorus levels in sediments of <250 mg/kg. Table 2.13-1 shows the reference streams along with poultry house density and total phosphorus in the sediment. Topographic data were used to calculate stream gradient and stream order to ensure that reference streams were similar in size and habitat conditions as the potentially impacted streams in IRW. The selection of reference streams were additionally based upon the results of sediment chemistry (typically total phosphorus <250 mg/kg), water quality (total phosphorus <0.030 mg/L), aerial topography, poultry house density in the watershed, and consultation with Oklahoma Department of Wildlife Conservation and Oklahoma Water Resource Board (Little Lee Creek and Spring Creek). Based upon the

sediment and water quality data and the low number of poultry houses in their respective watersheds, the following reference locations were selected (Figures 2.13-1, 2.13-1a and 2.13-1b):

- REF01/RS10003-Little Lee Creek- Oklahoma (Lee Creek Watershed)
- RS10004-Little Lee Creek-Oklahoma (Lee Creek Watershed)
- REF02- Dry Creek- Arkansas (Buffalo River Watershed)
- REF03- Spring Creek- Oklahoma (Neosho River Watershed)

The identified reference locations were sampled along with locations in the IRW during each of the major sampling programs conducted during the course of this project. During the initial river and biological sampling program in 2005, REF-01, REF-02, and REF-03 were sampled following identical sampling protocols and timeline as the sampling stations within the IRW. In 2006 and 2007, REF-01 and RS10004 were sampled during the 2006 and 2007 river sampling using identical sampling protocols and timeline as the sampling stations within the IRW.

As reported in Table 2.13-1 of Olsen (2008) the poultry house density (units of poultry houses per square mile in watershed) associated with each of the selected reference sites was as follows:

- REF01/RS10003: 0.14

- RS10004: 0.042

- REF02: <0.01

- REF03: 0.75.

In 2005, the specific sampling protocol used by CDM to collect fish at investigative and reference stations relied upon block nets at each end of the reach to prevent fish from immigrating or emigrating from the reach during collection. Fish samples were collected by electrofishing and seining within a 100-m reach at each location. Multiple passes (minimum of 2) were made within each reach using a combination of electrofishing and seining to deplete the fish available within the reach. Fish collected were kept in live wells until all fish collection was completed within a reach. Fish were identified in the field and recorded on field forms. Any abnormalities of the fish were to be noted. Fish not able to be identified in the field were preserved and sent back to the laboratory for identification.

In addition to the fish data collected, it was reported in the SOP 7-1 that other variables could be collected which may include but not be limited to:

- Average stream width, depth, and velocity with the sampling reach

- Water temperature, conductivity, pH, and dissolved oxygen content.

The stream characteristic data were provided on filed forms for the stations sampled in 2005, but the data were not available in Olsen's database. Water quality data were obtained from the laboratory reports.

Water quality data were not collected as part of the 2005 fish sampling event, but rather at other time periods during 2005.

The 2005 fish data used for our data analysis were obtained from Olsen's database dated April 19, 2008. No field forms were available to verify the database records, so the data were used as presented in the database.

2007 Fish Data—In 2007, 37 fish samples were collected from 35 stations within the IRW streams and an additional two reference sites located outside the IRW. As discussed above, as reported by Olsen (2008), the two reference stations were located along Little Lee Creek (i.e., RS10003 and RS10004). In 2007, the fish stations were sampled by CDM (includes investigative and reference stations) using either a backpack electroshocker, a boat mounted electroshocker, kick seining techniques, or a combination of these techniques (refer to SOP7-1.1 in Brown [2008]). Block netting was not used in 2007, but rather natural barriers or habitat types were used to define sample areas. A minimum 100-m and a maximum 800-m reach was sampled at each station. The actual reach length was to be defined as 30 times the average wetted width of the stream. Fish were collected from each habitat type within a reach, with a minimum of three pools and three runs sampled by electroshocking and three riffles by kick seining. The fish sampling was based on set time limits of effort for each habitat.

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The data collected at each sample location based on the SOP are as follows:

> At each fish sampling location, a variety of physical variables should be recorded in order to quantify factors that may have an influence on the resident fish populations and/or the efficacy of the sampling techniques employed. Variables may include, but are not limited to:
>
> - Average stream width, depth, and velocity within the sampling reach.
> - Amount and type of vegetation along each bank and in stream (e.g., 60% vegetated, primarily with grasses and shrubs)
> - Water temperature, conductivity, pH, and dissolved oxygen (DO) content.
> - Dominant substrate type and size for each of the four habitat types (pool, riffle, run/glide)
> - Numbers of each type of fish collected by unit of effort for each habitat unit (i.e., for each 3 minute unit of effort for a given pool or run and for each 30 second effort for each riffle).

As reported in SOP7-1.1:

> After each unit of time (initially set at 3 minutes), all fish captured will be either identified and counted or stored in a separate bucket or live well until the collection is complete for that habitat unit (e.g., a pool). The fish collected will be identified to the species level. Various fish identification manuals will be available including: *Fishes of Oklahoma* (Miller and Robinson 2004), *Fishes of Arkansas* (Robinson and Buchanan 1984), and the *Peterson Field Guide to Freshwater Fishes of North America* (Page and Burr 1991). Any specimen that cannot be positively identified in the field will be preserved and brought back to the lab for identification.
>
> All captured fish will also be observed for any physical abnormalities, and any findings will be recorded on the field data sheets. All fish population data will be recorded on the supplied data sheets using the fish species codes (Table 2 of SOP7-1.1). Any additional information relevant to this study will be recorded in field notebooks.

The majority of the fish samples were collected in July and August 2007. The total number of fish per taxa and incidence of lesions used for our data analysis were taken from files furnished from Dr. Jan Stevenson's computer hard drive (i.e., file Stevenson_Fish_Counts_071508) as a complete set of data for all 37 fish stations was not available elsewhere. The species counts available in these files were cross-checked against the available subset of the field forms. However, some species counts could not be checked because the field forms were not furnished

as part of Olsen's (2008) considered materials. Only minor differences between the hard drive files and the hard copy field forms were found, and most of the differences were a result of corrections in species names that were required between the field forms and the database. There was no information available to check the counts of lesions on fish, so these data were used as presented on the hard drive files of Dr. Stevenson.

Water quality measures of the streams were also collected in 2007, which included:

- pH

- Ammonia nitrogen

- Nitrate + nitrite (as N)

- Soluble reactive phosphorus

- Total dissolved phosphorus

- Total phosphorous

- Total organic carbon.

Water quality measures were collected in late winter or early spring of 2007 and then again in the summer of 2007 when the fish samples were collected. The data were obtained from the original laboratory electronic data deliverables that were provided.

Field parameters that were collected included:

- pH

- Dissolved oxygen

- Temperature

- Turbidity

- Conductivity.

Most of the field parameter data were collected in spring of 2007, much earlier than the fish were collected. The field parameters were obtained from field forms. No information on stream habitat conditions for 2007 was located in the files supplied by Stevenson or in Stevenson (2008). Apparently these habitat assessments were not conducted, which limits my ability to interpret the results, because habitat quality can dramatically affect the fish community structure within a reach. A habitat assessment is a basic component that should be conducted as part of a fish study (Barbour et al. 1999). Also, recent studies in both Oklahoma (Dauwalter et al. 2007) and Arkansas (Williams et al. 2003; Dekar and Magoulick 2007) have shown the importance of stream characteristics related to habitat to be major factors in determining the fish community composition. For example, Williams et al. (2003) showed that some of the key stream variables affecting the fish community composition were percent canopy cover, percent boulder substrate, percent cover of rooted vegetation, and bank stability. This paper also pointed out that fishes were influenced more by environmental variability that was unique to their historical sub-basins.

5.3.1.3   Spatial and Temporal Coverage

The 2005 fish sample stations are shown in Figure 5-2 and the 2007 fish sample stations are shown in Figure 5-16.

Table 5-7 provides sample locations for each station sampled in 2005 along with information about the sample location (i.e., stream name and sub-basin size). Within 2005 all samples were collected at stream stations reflecting medium to small sub-basins. No samples were collected at sample stations on the main stem of the Illinois River in 2005, which would reflect larger sub-basins (i.e., greater than 100 mi^2).

Table 5-8 provides sample locations for each station sampled in 2007 along with information about the sample location (i.e., stream name and sub-basin size). Most of the fish samples in 2007 were collected at sample stations representing medium sized sub-basins, but there were samples collected at stations representing small and large sub-basins as well, which provides a broad range of samples across stream size. Three samples were collected in the main stem of

the Illinois River, which represent very large sub-basins within the IRW (i.e., hundreds of square miles).

The BUMP sample stations monitored in the IRW are shown in Figure 5-17. A total of six BUMP stations are located within the IRW.

5.3.1.4   Taxonomic Levels

In both 2005 and 2007 the fish were identified to species level whenever possible. In certain cases the fish were reported as hybrid species of sunfish and so a specific species was not assigned. No distinction was made between adult fish and young of the year.

5.3.2   Summary of Fish Community

The fish metrics developed based on the 2005 and 2007 data are summarized by station in Tables 5-7 and 5-8, respectively. Common fish metrics that are used to describe the fish community were developed based on the fish data described previously. These fish metrics included:

- Taxa richness

- Total abundance

- Number of intolerant fish taxa

- Percentage of intolerant fish (of total fish abundance)

- Species diversity (as measured by the Shannon diversity index)

- Number of sunfish taxa

- IBI.

For 2005, the description of fish metrics is brief and provides only the range of the results for the ten Illinois River system stations. For the 2007 fish data, a more detailed description of the

fish metrics is provided because the data set was larger and additional statistical analysis of the data could be performed.

5.3.2.1   Taxa Richness

2005—In 2005, taxa richness varied between sample stations from a low of 10 species at station BS-208 to a high of 25 species at sample station BS-08. The minimum taxa richness was found in Peacheater Creek, which is a medium sized stream (sub-basin of approximately 22 mi^2). The highest taxa richness was found on Caney Creek, at a sample station (BS-08) representing the largest sub-basin (approximately 75 mi^2) sampled in 2005.

2007—In 2007, taxa richness varied between sample stations from a low of 5 species at stations RS-541 and RS-630 to a high of 30 species at sample station RS-757 (see Table 5-8 and Figure 5-18). The minimum taxa richness was found on an unnamed tributary of Tyner Creek and in Tahlequah Creek at stations that represented very small sub-basins. The highest taxa richness was found at a sample station on the main stem of the Illinois River, representing one of the largest sub-basins (659 mi^2) in the watershed. Taxa richness is affected by stream size (Thompson and Hunt 1930; Whiteside and McNatt 1972; Barila et al. 1981; Platts 1979; Beecher et al. 1988; Harrel et al. 1967), so the taxa richness data were evaluated in relation to sub-basin, which generally reflects the size of the stream. There was a significant statistical relationship (i.e., *P*-value < 0.0001) between fish taxa richness and sub-basin size (Table 5-9). The taxa richness of fishes was consistently higher in the Illinois River than in any of the other streams.

Taxa richness on the main stem of the Illinois River ranged from 25 to 30 species in the three samples collected within this water body. Species composition in the Illinois River included a variety of game fish species and forage fish at each of these sample locations (i.e., RS-757, RS-654, and RS-433A). Game fish included a variety of sunfish species (which include true sunfish, largemouth bass, smallmouth bass, and white and black crappie), channel and flathead catfish and yellow bullhead. Sunfish species were the dominant game fish and accounted for 40, 18, and 11 percent of the total fish abundance at stations RS-757, RS-654, and RS-433A,

respectively. At these same three Illinois River stations, bass species (including smallmouth, largemouth, spotted, and/or rock bass) accounted for 5, 3, and 5 percent of the total fish captured, respectively. The forage fish included a variety of shiner and darter species, with the dominant species being the cardinal shiner (see Figures 5-19 through 5-21). The cardinal shiner is a sensitive Oklahoma fish species as it is reported by Jester et al. (1992) to be "intolerant to degradation of habitat or water quality." This species is also classified as "sensitive" by the state of Kansas (KDWP 2004). As would be expected, the forage fish species comprised the dominant percentage of the fish, with the top trophic level game species making up a small percentage of the total fish. For example, at the three Illinois River sample stations, the percentage of smallmouth bass ranged from 3 percent at RS-433A to 0.1 percent at RS-654, which equates to a total of 19 and 1 smallmouth bass captured at each of these stations, respectively. Based on the sampling results at these three stations, there is a healthy and diverse population of game species inhabiting the Illinois River.

Only one sample was collected from a station outside the main stem of the Illinois River with a large sub-basin. This single fish sample was collected from a site on the Baron Fork river with a sub-basin of 305 mi^2. The sample contained 24 unique fish taxa, which is similar in magnitude to the taxa richness on the main stem of the Illinois River.

Stations with medium and small sub-basins had lower taxa richness. For example, the taxa richness at sample stations with medium-sized sub-basins (i.e., 20 to 100 mi^2) varied from a high of 20 species to a low of 11 species. At the sample stations with small sub-basins (i.e., less than 20 mi^2), taxa richness varied from a high of 21 species to a low of 5 species.

5.3.2.2   Diversity

Fish species diversity was estimated using the Shannon diversity index. As the species diversity or evenness increases, the Shannon diversity index increases. Species diversity is expected to decrease if there are stresses within a stream that reduce the number of sensitive species or result in the fish community being dominated by a particular species (uneven distribution of

individuals among species). Therefore, a high value of the Shannon diversity index is expected where lower stress occurs and more optimal fish habitat exists.

2005—In 2005, the Shannon diversity index varied between sample stations from a low of 1.19 at station BS-35 to a high of 1.92 at sample station BS-08. The minimum Shannon index was found on Fly Creek, at a sample station with a relatively small sub-basin (18 mi^2). The maximum Shannon index richness was found on Caney Creek, which was the largest stream sampled in 2005.

2007—In 2007, the Shannon diversity index varied between sample stations from a low of 1.01 at station RS-541 to a high of 2.58 at sample station RS-654 (see Table 5-8 and Figure 5-22). The minimum Shannon index value was found at a station on an unnamed tributary of Tyner Creek with a sub-basin of 7.16 mi^2. The maximum Shannon diversity index value was found on the main stem of the Illinois River at the Highway 62 crossing. Species diversity is affected by stream size (Whiteside and McNatt 1972; Barila et al. 1981; Platts 1979; Beecher et al. 1988; Harrel et al. 1967), so the Shannon diversity index data were evaluated in relation to sub-basin size to reflect the relative size of each stream at the location it was sampled. There was not a statistically significant relationship (*P*-value of 0.0202) between this species diversity index and sub-basin size considering the Bonferroni-adjusted significance level for multiple comparisons (i.e., *P*-value=0.0071; Table 5-9).

Although not statistically significant, species diversity was generally higher in the Illinois River than in any of the other streams, but species diversity was more variable within the range of stream sizes sampled than taxa richness.

The Shannon index on the main stem of the Illinois River ranged from 1.51 to 2.58 in the three samples collected within this water body. A single fish sample was collected on the Baron Fork, which had a large sub-basin (305 mi^2), similar to the Illinois River stations with a Shannon index value of 1.83, similar in magnitude to the Illinois River. The Shannon index values for sample stations with medium-sized sub-basins between 20 and 100 mi^2 varied from a high of 2.48 to a low of 1.53. At the sample stations with small sub-basins, Shannon index

values varied from a high of 2.10 species to a low of 1.02. Shannon index values at sample stations with very small sub-basins (i.e., less than 10 mi^2) varied widely (i.e., from 1.01 to 2.11).

5.3.2.3   Abundance

Fish abundance at each sample station was used an indicator of the health of the fish community. Fish abundance is expected to decrease if there are stresses within a stream that reduce the number of fish present.

2005—In 2005, fish abundance varied between sample stations from a low of 429 at station BS-208 to a high of 2,546 at sample station BS-68. The minimum fish abundance was found in Peacheater Creek at a sample station with a medium sized sub-basin (22 mi^2). The maximum fish abundance was found in Cincinnati Creek at a sampling station with a similar sub-basin (23 mi^2).

2007—In 2007, fish abundance varied between sample stations from a low of 114 fish collected at station RS-793 to a high of 1,151 fish collected at sample station RS-728 (see Table 5-8 and Figure 5-23). The minimum fish abundance was found on Shell Branch, which is a small stream that drains into the Baron Fork. The sample station was located near the headwaters of the stream, which is reflected by the very small sub-basin (4.2 mi^2). This fish station was located just downstream from the WWTP outfall for Westville Utility Authority. The maximum fish abundance was found in Caney Creek, at a stream station with a very small sub-basin (5.7 mi^2). Fish abundance is less affected by stream size but the fish abundance data were reviewed for consistency in relation to sub-basin. There was not a statistically significant relationship (P-value >0.05) between the fish abundance and the size of the sample station sub-basin. Fish abundance was generally higher in the Illinois River than the other streams, but fish abundance was more variable among different stream sizes that were evaluated than other fish metrics, such as taxa richness.

Fish abundance on the main stem of the Illinois River ranged from 629 to 843 in the three samples collected within this water body. A single fish sample was collected in Baron Fork, another large stream with a fish abundance value of 1,101, which is higher in magnitude

compared to the Illinois River. The fish abundance at the stream stations with medium sub-basins varied from a high of 822 to a low of 232. The fish abundance at the stream stations with small sub-basins varied from a high of 763 to a low of 287. The fish abundance at the stream stations with very small sub-basins varied from a high of 1,151 (maximum value among all stations) to a low of 114 (the minimum value among all stations).

Based on these data there was no general trend observed with fish abundance related with the size of the sample station sub-basin. Within the main stem of the Illinois River, fish abundance was consistently above the overall Illinois River system average fish abundance of 550. The results of the other stream orders were highly variable, with fish abundance values both much higher and lower than the average, indicating that particular habitat conditions at these streams stations may be more variable than on the main stem of the Illinois River.

5.3.2.4   Indicator Species

Indicator species within a fish community can provide insight into more subtle changes in fish communities than broader fish metrics like fish abundance. A common fish indicator species metric used is the number of intolerant fish taxa and percent of intolerant fish as a measure of the total fish abundance. Intolerant fish species are defined as those that are less tolerant to nutrient enrichment or other stressors (e.g., lack of a specific habitat requirement). Species were categorized as tolerant, moderately tolerant, moderately intolerant, and intolerant based on Jester et al. (1992), as recommended in Chapter 46 of the Oklahoma Water Quality Standards. Species are rated as either tolerant or intolerant to stress based on water quality and habitat. We used the water quality classification to determine the tolerance as the complaint was related to water quality issues. The moderately tolerant or moderately intolerant classifications were not counted when determining the number of tolerant and intolerant fish taxa. Tolerant fish taxa include such species as the red shiner, green sunfish, and black bullhead. Examples of intolerant fish species are the cardinal shiner, southern redbelly dace, and smallmouth bass. As the level of nutrient enrichment in a stream environment increases, the number of intolerant taxa and the relative percentage of intolerant fish are anticipated to decrease. Tolerance ratings are

generally non-specific to the type of stressor (Barbour et al. 1999), so are not necessarily related to water quality alone.

2005—In 2005, the number of intolerant fish taxa varied between sample stations and ranged from a low of five intolerant species at four stations (BS-117, BS-208, BS-35, BS-HF22) to a high of nine intolerant fish species at sample stations BS-HF04 and BS-08. The minimum number of intolerant fish taxa were found on the Illinois River, Peacheater Creek, Fly Creek, and Bush Creek at stream locations varying from very small to medium sized sub-basins. The highest number of intolerant fish taxa was found at Station BS-HF04 on Sager Creek, a sample station with a small sub-basin, and at station BS-08 located in Caney Creek, which had the largest sub-basin sampled in 2005.

2007—In 2007, the number of intolerant fish taxa varied between sample stations from a low of one intolerant species at stations RS-630 and RS-728 to a high of nine intolerant fish species at sample station RS-649 (see Table 5-8 and Figure 5-24). The minimum number of intolerant fish taxa was found at sample stations on Tahlequah and Caney creeks that have very small sub-basins (i.e., less than 10 mi^2). In addition, sample RS-728 on Caney Creek is located just downstream of the Stilwellada WWTP, and may be influenced by this outfall. The highest number of intolerant fish taxa was found at Station RS-649 on Baron Fork, which has a large sub-basin upstream from this station (305 mi^2). The number of intolerant fish taxa is affected by sub-basin size, so the numbers of intolerant fish taxa were evaluated in relation to sub-basin size. There was a statistically significant relationship (P-value = 0.0041) between the number of intolerant fish taxa and sub-basin size (Table 5-9).

The number of intolerant fish taxa on the main stem of the Illinois River (8, 8, and 7 taxa at stations RS-654, RS-443A, and RS-757, respectively) was consistently higher than the overall average of five intolerant species per fish sample. The maximum number of intolerant fish taxa species (i.e., nine) was found at a sample station along the Baron Fork with a sub-basin similar in size to the main stem Illinois River sample stations. The number of intolerant fish taxa at stream stations with medium sub-basins varied from a high of eight species to a low of three species. In streams with small sub-basins, the number of intolerant fish taxa varied from a high

of seven species to a low of three species. At the stream stations with very small sub-basins, the number of intolerant fish taxa ranged from a high of five species to a low of one species.

Another related factor that should be mentioned is that while the number of intolerant fish taxa varied dramatically from one to nine, the relative abundances of tolerant fish were always fairly low and ranged from 0 to 9.77 percent of the total fish catch at a given station. Therefore, the vast majority of fish in the IRW streams are not tolerant species, but rather less tolerant species at each sample station. For example, the three fish stations with the largest sub-basins located on the main stem of the Illinois River had relative abundance of tolerant fish equal to 0.16, 0.95, and 3.1 percent. The highest percent tolerance values (i.e., greater than 5 percent) were associated with sample stations that had smaller sub-basins (i.e., medium, small, or very small), but there was no clear trend within these three categories. In addition, there was not a statistically significant relationship (P-value > 0.05) between sub-basin size and the percentage of tolerant fish.

5.3.2.5   Sunfish Taxa Richness

Sunfish taxa richness was used as a relative barometer of the number of sport fish species found in each of the streams. The majority of the sport fish species present in the Illinois River system are sunfish; however, there are a few other species (channel and flathead catfish and yellow bullhead) that were observed primarily in the main stem of the Illinois, which are not reflected in sunfish taxa richness. Other fish species observed at sample stations other than catfish, bullhead, and sunfish species are not considered game fish (e.g., minnows and shiners).

The sunfish taxa include the sunfish (e.g., Redear, Longear, Bluegill, Green, and Warmouth), black and white and crappie, shadow bass, rock bass, spotted bass, smallmouth bass, and largemouth bass. Habitat for the sunfish species would be expected to be present more often in larger streams such as the Illinois River, where water depths are greater and physical condition of the stream provides suitable habitat for these larger fish. In smaller streams, habitat conditions needed for these game species are less likely to occur. For example, within the

smaller streams, spawning and feeding habitats for the sunfish species are less likely to be present because of the shallow depth of the streams.

2005—In 2005, the number of sunfish taxa varied between sample stations from a low of zero species at station BS-208 to a high of 6 sunfish species at sample stations BS-117 and BS-35. The minimum number of sunfish taxa was found in Peacheater Creek, at a sample station with a medium sized sub-basin (22 mi^2). The highest number of sunfish taxa was found in Fly Creek at a sample station with a small sub-basin, and in the Illinois River, at a sample station with a medium sub-basin.

2007—In 2007, the number of sunfish taxa varied between sample stations from a low of 0 species at stations RS-160, RS-541, RS-793, and RS-630 to a high of 11 sunfish species at sample station RS-757 (see Table 5-8 and Figure 5-25). The minimum number of sunfish taxa was found at stream stations located on Tahlequah Creek, Flint Creek, Shell Branch, and an unnamed tributary to Tyner Creek with small to very small sub-basins. In addition, one of these samples (RS-793) on Shell Branch is located just downstream of the Westville Utility Authority WWTP outfall, and may be influenced by this outfall. The highest number of sunfish taxa was found at station RS-757 on main stem of the Illinois River, which has a large sub-basin. The number of sunfish taxa can be affected by stream size, so the numbers of sunfish taxa were evaluated in relation to sub-basin size as an indicator of the stream size. There was a statistically significant relationship (*P*-value < 0.0001) between the number of sunfish taxa and sub-basin size (Table 5-9).

The number of sunfish taxa on the main stem of the Illinois River was consistently higher than the average number of sunfish species (i.e., three) and ranged from 6 to 11 species. A single fish sample was collected in Baron Fork, another large stream, where 8 sunfish species were observed, which was also well above the average for the IRW. The number of sunfish taxa found at sample stations with medium sub-basins varied from a high of six species to a low of one species. The number of sunfish taxa found at sample stations with small sub-basins varied from a high of four species to a low of zero species; those found at sample stations with very small sub-basins also varied from a high of four species to a low of zero species.

5.3.2.6   Index of Biotic Integrity

The fish IBI was calculated using a number of fish metrics to provide a more comprehensive barometric of the health of the fish community at each station. Fish IBI calculations were performed because they have regulatory significance in Oklahoma, because they are an important factor in determining if a stream is expected to support specific Fish and Wildlife Propagation beneficial uses or not, and whether a particular fish community will be supported by a stream. The majority of the streams within the IRW are classified as being within the cool water aquatic community subcategory of this beneficial use category. A single stream investigated within the IRW (i.e., Park Hill Branch) is classified as being within the warm water aquatic community subcategory. Different IBI score thresholds are used for each subcategory to determine if the subcategory (i.e., cool or warm water) of the Fish and Wildlife Propagation beneficial use is being attained or not (see further discussion later in this section).

The 2005 and 2007 fish data (from Olsen's IllinoisMaster.mdb and Stevenson's Stevenson_Fish_Counts_071508.xls, respectively) were used to calculate the fish IBI at each station within the Illinois River system and outside the IRW. The method for calculating the IBI was based on Title 785, Chapter 46, Implementation of Oklahoma's Water Quality Standards. Sample composition metrics were calculated at each station, which included:

- Total number of fish species (or taxa richness)

- Shannon diversity score

- Number of sunfish species

- Number of species comprising 75 percent of the sample abundance

- Number of tolerant species

- Number of intolerant species.

Species were categorized as tolerant, moderately tolerant, moderately intolerant, and intolerant based on Jester et al. (1992), as recommended in Chapter 46. The sub-basin size needed for

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scoring percent tolerant species, total number of species, and number of intolerant species was calculated for each sampling station with DEM using GIS.

The following fish condition metrics were calculated for each station:

- Percentage of lithophilic fish

- Percentage of deformities, eroded fins, lesions, and tumors (DELT) anomalies

- Number of individuals.

The categorization of fish as lithophilic or not was based on Dauwalter and Jackson (2004). For 2005 there was no DELT information available. For this reason, the assumption was made that there would be few to no fish lesions present at each station (i.e., IBI score of 5 for that parameter), based on the 2007 DELT results. In 2007 fish had a low percent of lesions and normally scored a 5 at most stations for this parameter.

Each sample composition and fish condition fish metric was categorized with a score of 1, 3, or 5 based on Appendix C of Chapter 46. Therefore, there is a maximum composition score of 30 (i.e., 6 factors times a factor of 5) and a maximum fish condition score of 15 (i.e., 3 factors times a factor of 5). The maximum overall score at each station is the sum of the all factors, which is 45. For the area of the state of Oklahoma that includes the IRW, the OWRB has different sets of IBI score limits to determine if the water body can support a warm water or a cool water aquatic community, respectively. These scores were based on data that had been collected on streams within Oklahoma by the state. The state has specific fish IBI score thresholds for the Boston Mountains and Ozark Highlands wadeable streams, which covers the geography of the IRW streams. For the warm water aquatic community subcategory, a stream with a score of 31 or greater is considered able to fully support a warm water aquatic community. A score at or below 22 indicates a surface water body that is not able to support a warm water aquatic community. Scores from 23 to 30 represent a surface water body with undetermined support for a warm water aquatic community. For the cool water aquatic community subcategory, a stream with a score of 37 or greater is considered able to fully

support a cool water aquatic community. A score at or below 29 indicates a surface water body that is not able to support a cool water aquatic community. Scores from 30 to 36 represent a surface water body with undetermined support for a cool water aquatic community. Surface water bodies with scores in the undetermined range require "additional investigation that considers stream order, habitat factors, and local reference streams" (OAC 785:46-15-5). The results of the final IBI score at each station are summarized in Tables 5-7 and 5-8.

2005—In 2005, the IBI scores varied between sample stations from a low score of 32 at station BS-208 to a high of 41 at sample station BS-08 (see Table 5-7 and Figure 5-26). The lowest IBI score was found in Peacheater Creek, at a sample station with a medium sub-basin. The highest IBI score was found on Caney Creek, at a sample station that had the largest sub-basin. All the streams sampled in the IRW in 2005 were classified under the cool water aquatic community subcategory, as were two of the reference sampling sites designated by the State of Oklahoma (BS-REF1 and BS-REF3). An additional reference site (BS-REF2) was located in the state of Arkansas, and therefore was not classified by the State of Oklahoma under either the warm or cool water aquatic community subcategory. Seven of the stream stations sampled in the IRW in 2005 had an IBI score of 37 or greater, indicating that they are predicted to fully support a cool water aquatic community (see Figure 5-26). Three of the stream stations sampled in the IRW had an IBI score in the range of 30 to 36, indicating that support of a cool water aquatic community is undetermined. No stations sampled in the IRW in 2005 were classified as not supporting a cool water aquatic community.

2007—In 2007, the IBI scores varied between sample stations from a low score of 21 at station RS-630 to a high of 45 (perfect score) at sample station RS-654 (see Table 5-8 and Figure 5-27). The lowest IBI score was found at a stream station located in Tahlequah Creek that had a very small sub-basin (i.e., 4 mi^2). The highest IBI score was found at Station RS-654 on the main stem of the Illinois River, which had the largest associated sub-basin (946 mi^2). Of the samples collected on the Illinois River, this station is located closest to Tenkiller Ferry Lake. The IBI score can be affected by stream size as measured by sub-basin size. For this reason, a number of the fish metrics that are used to develop the IBI score are dependent on sub-basin size.

Therefore, the IBI scores were evaluated in relation to sub-basin size. There was a statistically significant relationship (P-value = 0.0005) between the IBI score and sub-basin size (Table 5-9).

The IBI scores on the main stem of the Illinois River were consistently higher than the average IBI score (i.e., 34.9) and ranged from 39 to 45. A single fish sample was collected in on the Baron Fork at a sample station with a sub-basin size similar to the main stem Illinois River stations, which had an IBI score of 41, well above the average score. The IBI scores at stream stations with medium sub-basins varied from a high of 39 to a low of 33. IBI scores at stream stations with small sub-basins varied from a high of 43 to a low of 27, and at stream stations with very small sub-basins varied from a high of 41 to a low of 21.

All the streams sampled in 2007 were classified by the State of Oklahoma in the subcategory of cool water aquatic communities except for stream station RS-518 located on Park Hill Branch, which was classified as a warm water aquatic community (OAC 785:45). Figure 5-27 shows the IBI score by sample station in comparison to the Oklahoma category limits for determining if the stream can support a particular water aquatic community (cool or warm). Fifteen sampling stations were predicted to fully support a cool water aquatic community, and station RS-518 on Park Hill Branch was predicted to fully support a warm water aquatic community. Thirteen stations sampled within the IRW had an IBI score between 30 and 36, indicating that support of a cool water aquatic community was undetermined. Six stations sampled in the IRW in 2007 had IBI scores equal to or less than 29, and so were classified as not supporting a cool water aquatic community. The six stations included BS-35 (Fly Creek), RS-541 (on an unnamed tributary to Tyner Creek), RS-604 (on an unnamed tributary to Illinois River), RS-630 (Tahlequah Creek), RS-728 (Caney Creek), and RS-772 (on an unnamed tributary to the Illinois River). RS-728 is located just downstream of the Stilwellada WWTP outfall, and may be affected by this outfall.

Based on the IBI scores, the majority of the streams within the IRW are predicted to support a cool water aquatic community. In 2005, all of the stations sampled (100 percent) within the Illinois River system were predicted to fully support a cool water aquatic community or the status was undetermined. In 2007, approximately 17 percent of the stations (6 of 35) in the Illinois River were estimated to not support a cool water aquatic community. Their IBI scores

ranged from 21 to 29. Five of the six stations were located on streams with very small sub-basins. The sixth station had a small sub-basin. The majority of the stations sampled in 2007 in the IRW (83 percent) were either predicted to support a cool water aquatic community or the status was undetermined.

An IBI score is calculated using component scores for several factors: numbers of species, species diversity, number of intolerant species, species abundance, etc. Low IBI scores at the six stations listed above were a consequence of low numbers of species, low species diversity, low numbers of sunfish taxa, low numbers of species comprising 75 percent of the fish abundance at a station, and/or a low number of intolerant fish species. For these specific component scores, the values were 1 or 3 out of a maximum score of 5.

At all six stations with low IBI scores, the scores were high (score of 5 of 5) for percent of tolerant species, percent of lithophilic species (fish that spawn in stony benthos), and fish abundance. As described above, most of these sampling stations were in streams with very small sub-basins. Such streams may have very low or nonexistent flows during dry periods and do not normally support sport fisheries. However, each of these stations supported lithophilic species that are considered sensitive fish species. The four most dominant fish species at these six sample stations were lithophilic fish species, and included the cardinal shiner, orangethroat darter, stoneroller, banded sculpin, fantail darter, and southern redbelly dace. The cardinal shiner and southern redbelly dace are intolerant fish species, while the remaining two species are moderately intolerant. Other species common at some of these locations (and dominant at some stations) included the intolerant banded sculpin and stippled darter, and the moderately intolerant creek chub, fantail darter, and slender madtom. Thus, although the IBI scores were lower than 29 for the reasons presented above, the communities were nonetheless dominated by sensitive fish species. These results are not indicative of pollution effects, but are most likely the natural result of reduced habitat in the upstream areas of each stream, where the sub-basin size is limited.

5.3.3   Spatial Patterns of Community Characteristics

5.3.3.1   Relationship to Upstream Poultry Houses and Urban Land Use

I conducted an independent evaluation of the relationship between upstream poultry house density and fish metrics to determine if there was any relationship between the two factors. Upstream poultry house density was determined for each sampling station, and was used in place of the Plaintiffs' poultry house density metric. The methodology for deriving the Plaintiffs' poultry house density is unclear and appears to include poultry houses both upstream and downstream of sampling sites; it is unlikely that downstream poultry houses could substantially impact upstream sites.

The upstream poultry house density was calculated for each sample station by first defining the boundary of the upstream station-specific sub-basin within the IRW. The station-specific sub-basin is the area (in mi^2) that encompasses the potential flowing surface water of a sampling station (Table 5-10). The station-specific sub-basins for stream stations were delineated from 10-m DEM using the WATERSHED function of ArcGIS spatial analyst extension.

Data provided by the individual Defendants about the number of active poultry houses within the IRW were compiled and used to determine the number of active poultry houses within the station-specific sub-basin. The poultry house density for each sampling station was then calculated by dividing the total number of active poultry houses in the station-specific sub-basin by the station-specific sub-basin size.

In 2005, only 10 fish samples were collected within the IRW, and for the reasons stated previously, a full statistical analysis was not meaningful for this data set. In 2007, 35 sample stations located within the IRW were evaluated and provided a larger data set for statistical analysis. Stations outside the IRW were excluded from the statistical analysis. Both a linear regression analysis (see Table 5-9) and a nonparametric correlation analysis (i.e., Spearman rank) were performed on the 2007 data set (see Tables 5-11 through 5-13). The independent variables that were considered in the statistical analysis included poultry house density, sub-basin size, and percent urban land use. Sub-basin size was evaluated because it was

significantly related to a number of the fish metrics. Percent urban land use was evaluated because it was assumed to be an important factor in Dr. Stevenson's statistical analysis, and he used it to exclude a large proportion of the data set before performing his statistical analysis. Stevenson (2008) removed any sample stations from his statistical analysis that had a percent urban land use of 10 percent or greater.

Statistical analyses were used to evaluate relationships between each of the fish metrics and poultry house density, urban land use, and size of the sampling station sub-basin. Poultry house density was not related to any of the fish metrics for 2007 (Table 5-11 and Figures 5-28). Urban land use was not significantly related to any of the fish metrics either, except for a minor improvement in the linear regression model for number of intolerant taxa (Table 5-9 and Figures 5-29). Sub-basin size showed a significant relationship with a number of the fish metrics (Figure 5-30).

Based on linear regression models, sub-basin size was significantly related to the number of species (P<0.0001), Shannon diversity index (P=0.0202), number of sunfish taxa (P<0.0001), number of intolerant taxa (P<0.0001), and the total IBI score (P=0.0005). All of these measures increased with an increase in sub-basin size.

Spearman correlation analysis, a non-parametric method, confirmed the relationships quantified by the regression analysis. Number of species, number of sunfish taxa, number of intolerant taxa, and total IBI score all showed a statistically significant positive correlation with sub-basin size at a Bonferroni-adjusted significance level of 0.0071 (Table 5-13). The correlation with Shannon diversity index was not significant.

Based on the results of the 2007 fish sampling it can be concluded that sub-basin size is an important factor in the variability of the fish data between stream sampling stations within the Illinois River and its tributaries. However, CDM did not measure important habitat factors such as channel morphology, riparian vegetation, and sediment characteristics that may have important relationships with fish community variables. Poultry house density and percent urban land use have no statistically significant relationships with the structure of the fish communities within the IRW streams.

5.3.3.2   Relationships to Cargill Contract Growers and Breeder Operations

As discussed above, there is no overall relationship between upstream poultry house density and the structure of the fish community, as indicated by multiple metrics, within the Illinois River system. In addition, it is clear that there is no relationship between the presence of Cargill contract growers and breeder operations and the health of the fish community when fish sample stations that are located closest in the downstream direction from one or more Cargill contract growers are evaluated. Using the fish IBI as a barometer of the health of the fish community at a station, the stations located closest to Cargill contract growers were evaluated (see Table 5-14 and Figures 5-31 and 5-32). When more than one Cargill grower was located closest in a downstream direction to the station, these Cargill growers were grouped in Table 5-14. Station RS-757 along the main stem of the Illinois River had the greatest number of growers (15 farms) located upstream of the station and it had one of the highest IBI scores of 43.

The IBI scores for those closest stations located downstream of a Cargill contract grower or breeder operation ranged from 27 to 45. Ninety-two percent of the stations located closest in a downstream direction from one or more Cargill contract growers or breeder operations achieved an IBI score that classified them as either capable of fully supporting a cool water aquatic community (i.e., 37 or greater) or a score indicating that full support was undetermined (i.e., 30 to 36). This is higher than the overall average of 83 percent of stations achieving an IBI score of 30 or greater within the Illinois River and its tributaries. Only one station (BS-35) had a score predicting that a cool water aquatic community would not be supported.

The active poultry house densities at this subset of stations (stations closest in a downstream direction to Cargill contract growers) ranged from a minimum of 0.71 active poultry houses per mi^2 at Station RS-649 (below the average density of the Illinois River system, 1.34 active poultry houses per mi^2) to a maximum of 3.36 at Station BS-35 (see Table 5-14). However, station sub-basin size appeared to have a significant effect on fish community IBI scores. Stations with sub-basins less than 20 mi^2 (BS-35, RS-160, RS-399) exhibited the lowest IBI scores of the stations immediately downstream of Cargill contract growers (27, 33, and 31, respectively). As demonstrated in Section 5.3.3.1, these low scores are likely a result of small station sub-basins. All the stations located closest in the downstream direction to a Cargill

grower had an active poultry house density above the average within the Illinois River system, with the exception of RS-649 and RS-234. Even considering the high poultry house density at these stations, they represent the highest fish IBI scores collected within the Illinois River system. The single station with a value below 29 was located in a small sub-basin (17.87 mi^2).

Overall, stations located downstream of Cargill growers are supporting healthy cool water aquatic communities and reflect the general good condition of the fish community within the Illinois River and its tributaries.

5.3.3.3   Comparisons to OWRB Criteria for Beneficial Uses

The OWRB has different classifications of beneficial uses for surface water bodies (i.e., Fish and Wildlife Propagation, Aesthetics, Public & Private Water Supply, Agriculture, Primary Body Contact Recreation) for which they rate a water body and determine if it meets the requirements for attaining that beneficial use. The results of these evaluations are summarized on a yearly basis in a BUMP streams report. For purposes of evaluating the status of a watershed, the program selects stations that they monitor repeatedly on a yearly basis (or longer for some parameters) to gauge the health of the stream for the variety of beneficial uses. Within the Illinois River and its tributaries, there are six stations that are monitored as part of the BUMP (see Figure 5-17). At each of these stations, water quality is monitored and compared to numerical criteria such as dissolved oxygen concentration and turbidity, to evaluate whether criteria are being met for the beneficial use. There are also other biological criteria (e.g., the fish IBI score) that are used to evaluate whether attainment of the Fish and Wildlife Propagation beneficial use is occurring. If any one of the criteria is not met based on the requirements discussed in OAC 785:46-15-5, either based on water quality or biological criteria, a station is classified as not fully supporting the beneficial use of Fish and Wildlife Propagation.

The status of the six stations in the Illinois River system monitored as part of the BUMP for their ability to support the Fish and Wildlife Propagation beneficial use is provided in Table 5-15. Five of these six stations fully supported Fish and Wildlife Propagation over the past 5 (or 6) years for which BUMP reports were available (the stream report for 2006 is

currently unavailable). The one station where this beneficial use was not supported was located on the Illinois River (at US 59, Watts) and did not attain the status because of the turbidity of the water. Turbidity of the water has been a consistent problem at this station for the last 6 years, but the reason for the turbidity was not reported. There is no mention that this station did not fully support the Fish and Wildlife Propagation beneficial use because the fish community was affected.

In 2007, the BUMP Stream Report reported fish IBI scores for some of the stations collected in the Illinois River and its tributaries. Fish IBI scores were not available in other BUMP reports (2001 to 2005). The fish IBI scores by station are summarized in Table 5-16 and show that fish IBI scores ranged from 39 to 41 for the four stations where scores were reported. Therefore, all four of these areas along Baron Fork, Caney Creek, Flint Creek, and Sager Creek were determined to exceed the threshold of 37 and would fully support a cool water aquatic community. Note that although the fish IBI scores were reported in 2007, the data on which these scores were based were collected in 2003 or 2005, not 2007.

Based on the Plaintiffs' BUMP report, the Illinois River and its tributaries support Fish and Wildlife Propagation at most sample locations (i.e., 83 percent or 5 of 6), which is consistent with the results of the fish study conducted in 2007 and reported herein. The one station that did not attain this status was affected by a factor (i.e., turbidity) associated with general water quality, and not to a measured biological variable.

5.3.3.4   Comparison to EPA Region 6 Data

Another report of fish data that Stevenson did not consider was an evaluation conducted by EPA Region 6 in the Arkansas portion of the IRW. Although no areas of the IRW in Oklahoma were sampled, the report provides an important indication of upstream water quality of the Illinois River before it flows into Oklahoma. U.S. EPA (2004) evaluated the condition of the Illinois River system fish community at 10 sampling stations over a three-event sampling period (see Figure 5-33). They also collected samples at two reference stations, one located just north of the watershed and one in the watershed. Seven of these ten investigative stations showed no

impact to the fish community within the Arkansas portion of the IRW. This included two stations sampled near the Arkansas/Oklahoma border on the Illinois River. Looking more closely at the location of the three stations where some impact to the fish community occurred, two stations were sampled within urban areas and one was located just downstream of urban areas along Osage Creek. Osage Creek is a smaller stream, which, combined with the effects of urban land use, may be having an effect on the fish community at these sample locations. The EPA study considered habitat quality (unlike Stevenson) in both the riffle and pool environments. At two of the three sample stations where the fish community was considered impacted, the habitat quality was also impacted, which could be related to the urban environment. Therefore, the impact to fish at these stations is related to habitat quality and the effects of urbanization.

Based on the data collected by EPA in the Arkansas portion of the IRW, the fish community was impaired only in high urban areas; fish communities outside of urban-impacted areas were determined to be healthy, diverse, and abundant.

5.3.4   Summary

Based on the available fish data collected in the Illinois River and its tributaries, the watershed supports a healthy fish community. A number of fish metrics were estimated based on the 2005 and 2007 fish data to evaluate the health of the fish community, including taxa richness, diversity, abundance, number of intolerant taxa, number of sunfish taxa, and the fish IBI. Taxa richness and diversity of fish (including game fish species) within the Illinois River system is greatest within the main stem of the Illinois River, which is the largest flowing stream within the IRW, with the largest sub-basin in the watershed. Within the main stem of the Illinois River, as many as 30 species of fish were collected at a single station, including many game fish species. The number of fish species and a number of the other fish metrics were generally lower in streams with smaller sub-basins. Fish community changes that occur among stream stations (e.g., number of fish species) within the IRW appear to primarily relate to sub-basin size (i.e., roughly speaking, the size of the stream) based on statistical analysis of the 2007 fish data. These types of effects on fish communities are well documented.

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A statistical analysis of the 2007 data was performed to determine if there was any relationship between urban land use and poultry house density, which had been the basis of Stevenson's statistical models for each fish metric. In addition, sub-basin size was evaluated in the analysis to control for urban land use, which has been well documented to affect the fish community. My own independent statistical analysis of the fish data showed that there was no statistically significant relationship between fish metrics and urban land use or poultry house density. My analysis did show a statistically significant relationship between sub-basin size and many of the fish community metrics. The variation in the fish metrics from the smallest to largest sub-basin sampled within the IRW varied on average by 51 percent, with the greatest percent change of 83 percent.

Lastly, a review of fish data collected by the State and by EPA within the IRW was conducted. Based on these data, the Illinois River and its tributaries support a cool water aquatic community. Most stations evaluated by the state (83 percent) either fully supported a cool water aquatic community or were in a range where full support was undetermined (OAC 785:46-15-5). EPA found the fish community to be unimpacted at 70 percent of sample stations (U.S. EPA 2004). From the review of the stations where the fish community was not fully supported or was somewhat impacted, it appeared that other environmental factors within the IRW were the cause rather than poultry-related operations. Taking into account the available fish data collected within the Illinois River and its tributaries, this system supports a healthy fishery in most areas and there are no effects related to poultry house density.

6   Evaluation of Stevenson (2008)

6.1   Overall Approach

With regard to alleged injuries to biological resources in the streams of the IRW, Dr. Stevenson analyzes three particular aquatic communities (benthic algae, BMI, and fish) within the streams to evaluate the potential effects of nutrient loading from poultry operations on the streams. He attempts to establish a relationship between increased phosphorous in surface waters of streams of the IRW allegedly associated with poultry operations by using an estimate of poultry house density at each station sampled within the Illinois River and its tributaries. Poultry house density is the primary independent variable that Dr. Stevenson used as an indicator of the amount of nutrient loading allegedly associated with poultry operations that will occur to a stream in the Illinois River and its tributaries. He evaluates the relationship between phosphorous concentrations and surface water, along with other surface water quality measures, such as dissolved oxygen and pH, to try to make a linkage between poultry house density and nutrient loading to the streams. The poultry house density is then related to specific measures of the biodiversity of the stream algae, BMI, and fish community to evaluate whether or not there is an effect on these aquatic communities related to poultry house density. The following is the general evaluation process that Dr. Stevenson used to evaluate whether the benthic algae, benthic invertebrate, or fish community was being affected by poultry operations as reported in Stevenson (2008).

1. Surface water quality data were monitored at a number of stations on the streams in the IRW. In addition, an estimate of poultry house density was calculated based on Engel (2008) and Fisher (2008) data at each of these sample stations. Stevenson used statistical analysis methods in an attempt to establish a relationship between poultry operations and nutrient loading to the stream at specific sampling stations. The collection of these data occurred one or more times over the study period.

2. At varying numbers of the sample locations monitored for surface water quality, samples of algae, BMI, and fish were collected. These algae, BMI, and fish data were used to derive metrics (e.g., measure of species diversity) that were used as indicators of the health of each of these aquatic communities.

3. Dr. Stevenson used statistical analysis methods to evaluate whether or not there were statistically significant relationships between the aquatic community metrics and the poultry house density or measure of stream surface water quality (e.g., total phosphorus). He used his statistical models to estimate amounts of change in specific metrics in a haphazard way to predict changes in the aquatic community in specific cases. For example, he used the relative percent change in fish metrics to make a prediction of injury specifically to the fish community within the IRW.

Although Dr. Stevenson does not explicitly claim to be conducting a NRDA, his report (Stevenson 2008) uses a key regulatory term that is important in such assessments (i.e., "injury"). Because Count 2 of the complaint for this matter concerns alleged natural resource damages under the Comprehensive Environmental Response, Compensation and Liability Act of 1980 (CERCLA), it is important to evaluate the degree to which the assessments of Stevenson (2008) comply with the general concepts and assessments that are part of a CERCLA NRDA. I will address three important aspects of a natural resource damages (NRD) claim relative to Stevenson (2008):

1. Have baseline conditions been established that can be used as a comparative basis for quantifying any injury to natural resources?

2. Have causal relationships been demonstrated between the release of a hazardous substance and any injuries to natural resources?

3. Have natural resources in streams of the IRW been shown to be injured by any releases of hazardous substances?

My assessment of these three aspects of an NRD claim is focused primarily on the BMI and the fish communities of the IRW. The claims associated with effects on the algae community are primarily based on arguments of aesthetics, which are outside the scope of this report.

6.2   Statistical Approaches

6.2.1   Quantification of Poultry House Density Effects

As will be discussed in subsequent sections, Dr. Stevenson's statistical evaluations of the relationships of many biological variables to poultry house density is fundamentally flawed because it does not represent a density of poultry houses that could theoretically be contributing nutrients from litter application to the stream sampling stations, and because the data set was inappropriately censored and otherwise transformed to confound meaningful analyses. In addition to these fundamental errors in logical design of the analyses, Dr. Stevenson also made extensive errors and inconsistent applications of statistical techniques.

Throughout Stevenson (2008), the magnitude of effects attributable to poultry house density was estimated as the relative percent change in predicted value from the minimum to the maximum poultry house densities observed, with all other variables set to their respective median values. This calculation is applied to all of the fish response variables in Table 4.2 of Stevenson (2008), including those unrelated to poultry house density (note that Table 4.2 excludes the proportion of lithophilic individuals with no explanation). The conclusion of 20 percent loss across the range of poultry house densities observed is calculated as the average percent change across all of the fish response variables. This average is not meaningful because it represents the average of the maximum change for each of the 13 variables, 12 of which show no significant relationship with poultry house density.

Table 4.1 of Stevenson (2008) summarizes the array of regression models fit to each fish metric, totaling 52 models (13 metrics fit to four independent variables). Significance in Table 4.1 appears to be determined at a 0.10 significance level rather than the more conventional 0.05 significance level used throughout the majority of Dr. Stevenson's report. Further, no

adjustment was made to account for the number of statistical models that were analyzed. The Bonferroni adjustment is one of many methods, and one Dr. Stevenson uses elsewhere in his report, that adjusts the significance level to account for the number of assessments being conducted (Piegorsch and Bailer 1997). For example, if the same model were fit to 20 different variables, using an unadjusted 0.05 significance level, one would expect to find one model statistically significant by chance alone ($0.05 \times 20 = 1$) even if no relationships actually existed. Dr. Stevenson applied a Bonferroni adjustment to the results of models fit to the diatom metrics (Table 3.2 of Stevenson 2008), but apparently did not use it with the models fit to the fish metrics.

Only four of Dr. Stevenson's models fit to the poultry house density are indicated as significant in Table 4.1 of Stevenson (2008). Two of these relationships are no longer significant when the more conventional 0.05 level is applied, and only one of those remaining models would be considered significant after a Bonferroni adjustment was applied to the significance level to account for the 13 models ($0.05/13 = 0.0038$ adjusted significance level). On this basis, only the number of lithophilic taxa is considered statistically significant related to poultry house density based on Stevenson's overall approach as modified above.

Moreover, nowhere within Stevenson (2008) is the uncertainty or variability of his statistical predictions addressed. Even when derived from statistically significant models, predictions made at the extreme ranges of the independent variables (such as poultry house density) have the highest uncertainty associated with them, and as a result the widest confidence intervals. A confidence interval represents the range over which the actual value being predicted could likely fall with the specified confidence level. Frequently, a 95 percent confidence level is used to mimic the 0.05 significance level. Most of the figures included in Stevenson (2008) that show two variables with a fitted regression line also indicate the confidence interval around the fitted line (Figures 2.8–2.11, 2.13–2.15, 2.24, 2.28, 2.31–2.32, 2.34–2.37, 2.40–2.41, Figures 3.1–3.3 of Stevenson 2008). These figures clearly show increased uncertainty at the minimum and maximum poultry house density values (confidence intervals are wider at the ends of the range than in the center). This indicates that Dr. Stevenson was aware of the uncertainty associated

with predictions at the extreme ranges of the data. However, he neglected this important information in his calculation of relative percent change for each metric.

Because of the lack of complete details included in his report and production files, repetition of his analyses to obtain the confidence intervals for the predicted poultry house effects was not feasible.

6.2.2   Inconsistent Reporting of Results

Although the general analysis approach used in Stevenson (2008) is consistent for algae, invertebrate, and fish response variables, the summaries of findings and associated results are inconsistent. For example, Table 2.2 (algal biomass) of Stevenson (2008) summarizes correlations as significant based on a 0.05 level for 2-sided comparisons with no adjustment for multiple comparisons. Table 3.2 (diatom metrics) of Stevenson (2008) summarizes significant correlations based on one-tailed comparisons at a 0.05 significance level after a Bonferroni adjustment. Table 3.3 (diatom species composition) of Stevenson (2008) shows no indication of which relationships were considered significant. Tables 3.4, 3.5, 3.6, and 3.7 of Stevenson (2008) indicate significance without mentioning the level used or any adjustments. These tables also indicate significant relationships that are the reverse of expected impacts. Table 4.1 (fish responses) of Stevenson (2008) indicates significant relationships at a 0.10 level with no multiple comparison adjustment. Therefore, the statistical significance results presented in Stevenson (2008) are an inconsistent compilation of different approaches. Given the nature of the data analyzed by Dr. Stevenson, the most appropriate approach would be to use two-sided comparisons at an overall 0.05 significance level after an adjustment for the number of comparisons. Without an *a priori* expectation of the direction of change in the tested variable, a two-sided test is appropriate. Moreover, the 0.05 significance level has been generally accepted as a standard of practice in ecological assessments. Based on the results tabulated within his report, Stevenson did not use this approach on any of his results. All of the regression models summarized determined significance of variables statistically (i.e. based on the coefficient significance level or *P*-value), and made no assessment of the biological significance of the relationships. Given a large enough sample, statistical significance can be achieved at levels far

too small to be biologically meaningful. Further, many of the regression models summarized include additional covariates that may not be significant. Many of the summary tables do not include significance levels for all of the parameters analyzed within the models. The variability attributed to these additional parameters may decrease the residual error term, thus making significance of individual coefficients more easily obtained. Many of the regression models with significant parameters still explain only a small amount of the overall variability. For example, poultry house density explained generally less than 30 percent of the variability in the diatom indicator variables (Table 3.3 of Stevenson 2008). Predictions from models explaining this low amount of variability may be deceiving without also reporting the associated confidence range to quantify the plausible range of values.

6.2.3   Inappropriate Use of Data Transformations

Dr. Stevenson applied data transformations to most biological variables assessed in order to meet the assumption of normality that underlies both the correlation and linear regression analysis methods used. The variety and abundance of transformations used by Stevenson is unconventional and the resulting analyses are therefore difficult to interpret. For example, it is difficult to understand how to interpret relationships with the square-root of the square-root of the proportion of Trichoptera; or the square-root of the square-root of the proportion of shredder taxa (Table 3.1 of Stevenson 2008). Multiple transformations are not commonly used except in specific instances (arcsine square-root transformation of proportion data). In using these unconventional transformations, Dr. Stevenson provides no scientific rationale or cited references that would justify his approaches. In addition, it appears that no systematic approach was used to determine what transformations were applied to which variables, with multiple transformations applied successively until normality could be achieved.

Transformations should be selected to meet the underlying assumptions of the analysis method (normality and homogeneity of variance) but should also be motivated by the scientific meaning of the resulting variables. Similar metrics should logically receive the same transformation, making results directly comparable. For example, the Box-Cox or power transformation (Box and Cox 1964) can be used to recommend an appropriate transformation to achieve normality

and stabilize variance (Piegorsch and Bailer 1997). Moreover, Dr. Stevenson's final results based on the transformed variables were never compared with parallel non-parametric analyses or analyses applied to non-transformed variables to confirm that the transformations were not altering conclusions.

In summary, the statistical analyses reported in Stevenson (2008) are inconsistent and scientifically flawed. These deficiencies, when combined with the underlying flaws in his overall methodology, result in invalid and scientifically meaningless results.

6.3   Inappropriate Characterization and Selection of Reference Stations

Comparing site-related conditions with appropriate reference areas is an accepted and well-documented approach that is used in NRDAs (i.e., for definition of baseline), ERAs, and other environmental assessments. In conducting such comparisons, it is essential that the reference area(s) be similar to the assessment area (i.e., the IRW streams) except for the magnitude of the stressor being assessed. In other words, in the case of the Illinois River and its tributaries, the reference area stations selected for sampling should have included all other potential influences, natural and anthropogenic, on biological communities except for the releases of phosphorus or other constituents from poultry litter application sites. These reference stations would then be compared rigorously to the investigative sample stations (i.e., stream stations potentially influenced by poultry operations). These reference condition comparisons are an important element of the studies conducted to evaluate potential effects of poultry operations on benthic algae, BMI, and fish communities within the Illinois River and its tributaries.

Dr. Stevenson acknowledges that "reference condition" has regulatory significance and then defines this term as "[r]eference condition is the physical, chemical, and biological condition found in streams having watersheds with the lowest level of human activities." This is a fundamental error when comparisons to a reference area are being used to determine whether a particular area is being adversely affected by a particular stressor. The fundamental problem with this approach is that nutrient enrichment can occur from many human influences other than

poultry operations, including but not limited to urban land use practices and other agricultural practices. For the IRW streams, many reference stations would be required to accurately characterize the varied conditions at the investigative stream stations that are unrelated to poultry operations. These reference stations would have similar habitat and water quality characteristics to the investigative stream stations without any poultry litter application. The reference stations should bracket the IRW investigation stream stations for characteristics such as the following:

- Full range of stream sizes as reflected by sub-basin size and general hydrological characteristics

- Degree of urban development and stream habitat modifications

- Sewage discharges

- Other nutrient sources such as agricultural operations, grazing, septic tanks, and plant nurseries.

As a key example of the limitations of Dr. Stevenson's approach, as part of the 2007 fish study, only two reference stations were selected to characterize the conditions of the 35 investigative sample stations. These two reference stations were located on Lee Creek outside of the IRW. These stations are on located within 10 miles of each other, and had medium-sized sub-basins in the same stream reach. This does not reflect the varied conditions within the IRW streams. Within the Illinois River and its tributaries, investigative fish samples were collected from a wide range of stream sites. These sampling stations represented a range of locations, from very small sub-basins to stations within the main channel of the Illinois River. This provides one concrete example of the gross inadequacy of the reference areas selection. More detailed discussion of factors important to selecting reference stations are discussed in more detail in Section 6.6 (BMI) and 6.7 (fish), respectively.

In NRDAs, reference areas are frequently used to establish the baseline conditions and services. Conditions at the site being assessed are then rigorously compared with baseline to determine if there are site-related injuries to resources, and to estimate any service losses. The reason that

reference areas must be carefully selected when conducting ERAs or NRDAs is that the use of inappropriate reference areas can lead to the "ecological fallacy." As discussed in Suter (1993), "[i]n ecological assessments, the 'ecological fallacy' occurs when populations and communities found in association with pollution are compared with populations and communities at less polluted sites, and any biological differences are attributed to the pollution." This situation can occur where natural differences in habitat quality, natural variability of biological communities, and influence of other anthropogenic stressors exist between the reference and assessment areas. Then, when comparisons are made, any apparent differences are attributed incorrectly to effects of substances at the more contaminated site. In such situations, the magnitude of the ecological fallacy can be compounded by the application of inappropriate statistical methods (as referred to Section 6.3) and by not having a rigorous framework for evaluation of causal relationships.

Dr. Stevenson's analysis approach claims to evaluate impacts to the Illinois River and its tributaries through a causal pathway analysis, but this approach is flawed because he never adequately characterizes reference conditions. His *a priori* approach begins by evaluating urban land use and poultry house density impacts on total phosphorus concentrations, the stressor he believes to be the cause of the problems related to poultry house density. Dr. Stevenson cannot substantiate this causal pathway because he never first adequately characterizes reference conditions and the concentrations of phosphorous related to those reference conditions. This is a fatal flaw in each of his biological evaluations.

In summary, Dr. Stevenson uses an incorrect definition of reference area and therefore reaches erroneous conclusions concerning any comparisons of the biota of IRW streams with the putative reference streams.

6.4   Relationship of BMI Communities to Stream Characteristics, Phosphorus, and Poultry House Density

As part of his expert report, Dr. Stevenson evaluated the BMI community in the Illinois River system in relation to assumed impacts of poultry house density. The objectives of this analysis were to demonstrate:

- The number of reference taxa and individuals sensitive to pollution will decrease with increasing poultry house density in watersheds, nutrient concentrations, and nutrient-related stressors in streams;

- The number of taxa and individuals that are tolerant to pollution will increase with increasing poultry house density in watersheds, nutrient concentrations, and nutrient-related stressors in streams.

- The trophic structure of invertebrate assemblages will change with increasing poultry house density in watersheds, nutrient concentrations, and nutrient-related stressors in streams (Stevenson 2008).

However, inappropriate sampling methodology (including lack of suitable reference sites and a deficiency of sampling site habitat information) and flawed statistical analyses severely limit any meaningful interpretation of these results. My main criticisms of Stevenson's evaluation of BMI community health are encompassed in the following categories:

- Improper definition of baseline conditions

- Inappropriate selection and use of reference sites

- Lack of consideration of stream habitat characteristics

- Flawed interpretation of BMI results.

Each of these key problems in his evaluation of the BMI community of the Illinois River system is discussed further below.

6.4.1   Improper Definition of Baseline Conditions

BMI community structure naturally varies over habitat gradients. Even seemingly small changes in physical habitat characteristics, including substrate size, riparian vegetation, current, and bank stability, can exert a substantial impact on the composition of BMI communities (Stark 1993). If habitat characteristics are not accounted for in a biological assessment, natural community variability can be mistakenly ascribed to changes in water quality (Lenat and Barbour 1993).

During a biological assessment, natural community variability can be accounted for by 1) the use of sufficient and appropriate reference sites and 2) the definition of sampling site habitat variability through standardized habitat assessment techniques (Reynoldson et al. 1997). Neither of these methods was implemented during benthic surveys conducted by CDM in 2006 and 2007, indicating that natural variability in benthic communities may have been improperly identified as an injury and incorrectly attributed to poultry house density.

6.4.2   Inappropriate Selection of Reference Sites

Within a biological assessment, reference sites are used to estimate baseline BMI community variability in the absence of stressors of interest. The reference sites used in 2006 and 2007 (RS-10003 and RS-10004) were located in the same stream system (Little Lee Creek), within 10 miles of each other (Figures 5-3 and 5-4). As such, the benthic community and habitat variation between these two sites will likely be minimized. The IRW, on the other hand, covers more than 1,500 mi^2, and contains numerous tributaries; Illinois River system benthic communities surveyed in 2006 and 2007 were located at sites ranging from small headwater streams to stations in the main stem of the Illinois River. Multiple reference sites, covering a diverse and heterogeneous landscape, are necessary to develop a baseline for a large regional bioassessment such as was conducted here (Hughes et al. 1993). Reliance on a minimal number of control sites is problematic because this approach provides a limited estimate of natural variance and a poor extrapolation to study sites (Reynoldson et al. 1997). Current standards suggest the paired comparison of study and reference sites based on similarity of habitat and land characteristics or the use of many reference sites so that the entire variability of the assessment area can be covered by a "reference envelope."

6.4.3   Lack of Consideration of Stream Habitat

Variability in land use, localized limnology, riparian vegetation, stream substrate, and hydraulic properties can strongly affect the community composition of biota, including BMI (Crunkilton and Duchrow 1991; Hawkins et al. 2000; Stark 1993; Vinson and Hawkins 1998). Localized substrate characteristics are particularly important drivers of benthic community structure. For

example, biodiversity and abundance of benthic communities may be greatest in riffle areas with pebble-sized substrate (diameter ~20mm), whereas less diverse and abundant BMI communities are usually collected from sandy substrates (Erman and Erman 1984). Substrate interstitial space and porosity are also significant determinants of benthic community structure and diversity (Duan et al. 2008). Even in agricultural catchments with both physical stressors and nutrient enrichment, substrate characteristics and riparian cover were shown to have a more significant influence on BMI community composition than either nitrogen or phosphorous concentrations (Richards et al. 1993).

In order to properly conduct a bioassessment of benthic assemblages, a thorough evaluation of physical and habitat variability must be performed, and the results of this used to inform both the study design and the data analysis (Norris and Georges 1993). Although habitat assessments were performed in conjunction with preliminary 2005 sampling, these assessments were not performed in subsequent sampling efforts in 2006 and 2007. As a result, it is impossible to gauge the impact of habitat structure on Illinois River system benthic communities, or to separate the relative influence of physical variability from nutrient effects. Without specific information regarding physical variables such as substrate composition at each sampling site, it is unclear how much of the variability of sampled macroinvertebrate communities could be explained by natural habitat variability. Therefore, any comparisons of benthic community structure among sampling areas would be semi-quantitative at best, and are not reliable indicators for assessing the effects of any specific habitat factor or stressor.

6.4.4   Flawed Interpretation of the BMI Results

Dr. Stevenson concluded that nutrient enrichment was impacting BMI communities based solely on the benthic data collected in 2007. For 2006 BMI data, Stevenson (2008) concluded that "relatively few indicators of species composition were related to stressors . . . [and the] response was opposite of the usually predicted response for pollution." However, his analysis and interpretations of the 2007 BMI data are profoundly flawed, both in terms of nutrient indicators used and benthic metric selection and analysis.

First, it is unclear from the text whether the Bonferroni adjustment method was applied during Dr. Stevenson's analysis of BMI data. The Bonferroni adjustment is necessary to correct the significance level when a number of assessments are conducted on the same data set, as was done here. Analyzing data without applying this adjustment would lead one to identify insignificant relationships as significant. A more detailed discussion of the importance of this is presented in Section 6.2.1. Further, the reduction of the total Illinois River system data set to a "low-urban sites" data set was an unnecessary component of the analysis that most likely resulted in flawed conclusions. Dr. Stevenson arbitrarily removed sites with greater than 10 percent urban land use, which created a non-meaningful and probably biased subset of data. Finally, the use of algal growth metrics, pH, and dissolved oxygen as indicators of nutrient enrichment were not adequately justified by Dr. Stevenson. His failure to establish a causal pathway between poultry house density and algal metrics, pH, and dissolved oxygen is discussed in depth in Section 6.2.3. Briefly, no legitimate, statistically-relevant relationships were demonstrated in Stevenson (2008) between poultry house density and these indicators, as a result of improper multiple transformations and questionable use of significance levels greater than 0.05. Therefore, it is inappropriate to cite relationships between these unverified nutrient indicators and benthic metrics as confirmation of adverse effects of nutrients on BMI communities. As a result, square root of the percent of filamentous green algae cover (SPCGREENCOV), standard deviation of dissolved oxygen concentrations (STDEVDO), maximum pH (MAXPH), and minimum dissolved oxygen concentration (MINDO) will not be considered herein as appropriate nutrient indicator variables.

Six of the 17 utilized species composition metrics listed in Table 3.5 of Stevenson (2008) were not referenced to a particular scientific source (invertebrate taxa richness, proportion of insects, proportion of EPT, proportion of Trichoptera, proportion of Ephemeroptera, and proportion Chironomid). Barbour et al. (1999) identifies two of these as best candidate benthic metrics for gauging community health: proportion EPT and proportion Ephemeroptera, along with a third metric listed in Table 3.5 of Stevenson (2008), percent tolerant individuals. None of these metrics was significantly correlated with total phosphorus concentrations. In fact, there were no significant correlations between total phosphorus and any of the benthic metrics selected by Dr. Stevenson when the complete data set was analyzed.

The majority of Dr. Stevenson's functional feeding group metrics appear to have been inappropriately attributed to Barbour et al. (1999), and consequently there is no scientific justification for their use. Only two of the 14 attributed functional feeding group metrics in Table 3.1 of Stevenson (2008) were correctly referenced—proportion of filter-collector individuals (SQRTPROPFC) and proportion of gatherer-collector individuals (PROPGC). The remaining 12 indicators, including the two functional feeding indicators deemed significant in Table 3.7, were not defined in Barbour et al. (1999) as reliable benthic indicators of perturbation. Relative abundances of invertebrate functional feeding groups (e.g., as proportions or percentages of total abundance) were recommended indicators of benthic community health in this reference. However, Dr. Stevenson used functional feeding group taxa richness instead of functional feeding group abundance. As a result, there is no reliable documentation provided for the predicted responses of these metrics, and it is therefore impossible to assess Illinois River system BMI community health based on functional feeding group taxa richness. However, neither the relative abundance of filter-collectors or gatherer-collectors (SQRTPROPFC and PROPGC) showed a significant correlation with total phosphorus concentrations.

Additionally, even a cursory appraisal of BMI community structure across the three sampling years would indicate that communities sampled in 2005 and 2006 are different from those sampled in 2007. Most significantly, relative abundance of chironomid larvae increased by 4- to 6-fold in the spring of 2007 versus the summer of earlier years. Dr. Stevenson attributed this year-over-year difference in community structure to effects of nutrient enrichment:

> The strongest effects of nutrient pollution on macrobenthic invertebrate communities were in spring 2007 compared to summer 2006. . . [and] Trichoptera responded to nutrients in one season but not in the other (Stevenson 2008).

This conclusion fails to account for the strong, well-documented and predictable oscillations in BMI community abundance and composition with season (Bêche et al 2006; Linke et al 1999; Murphy and Giller 2000). Seasonal fluctuations in precipitation and temperature result in predictable changes in stream habitat condition and food availability; aquatic invertebrate life cycles are often finely attuned to these changes. Sampling events in 2007 occurred in April, as

compared with the late-summer/fall surveys in 2005 and 2006, which would likely explain why the 2007 BMI community composition is unique. Additionally, a comparison of 2006 and 2007 reference communities presents an even more compelling argument that seasonal BMI fluctuations confounded the conclusions presented in the Stevenson expert report. As with Illinois River system benthic communities, 2007 reference communities contained about four times the relative dipteran abundance of 2006 BMI samples, indicating that the spring increase in dipteran abundance was occurring naturally in the area.

The lack of consistent correlation of BMI metrics to nutrient concentrations and "related stressors" over 2006 and 2007 should have been a strong indication that poultry house density had no significant impact on benthic community health. Instead, this lack of correlation was attributed to "the potential variability in responses of species . . . [and that] most invertebrate identifications are to the genus level . . . [creating a] loss of finer information for invertebrates" (Stevenson 2008). In fact, accurate species-level keys and descriptions are not available for many BMI species, and family- and genus-level identifications are commonplace in BMI bioassessments (Resh and McElravy 1993). It is not reasonable to assume that 1) BMI impacts seen only during the 2007 sampling season are a result of nutrient contamination, and that 2) taxonomic refinements would dramatically alter 2006 results to show that there had been injury. This ignores obvious seasonal effects apparent in both reference and Illinois River system benthic communities.

6.5   Relationship of Fish Communities to Stream Characteristics, Phosphorus, and Poultry House Density

Within his expert report, Stevenson (2008) evaluates the fish community to determine if there is any effect on the fish community associated with poultry house density. As stated in Stevenson (2008):

> The objectives of this section of the report are to document the injuries of fish species composition that are related to poultry house activities and nutrient pollution. I used the species composition of fish to calculate indicators of biological condition to measure injury. I hypothesize:

6-15

The number of taxa and individuals sensitive to pollution will decrease with increasing poultry house density in watersheds, nutrient concentrations, and nutrient-related stressors in streams;

The number of taxa and individuals that are tolerant to pollution will increase with increasing poultry house density in watersheds, nutrient concentrations, and nutrient-related stressors in streams.

While Stevenson starts with these focused objectives in mind (i.e., two basic fish metrics), he uses flawed statistical techniques and assumptions to produce an analysis of the fish community that is scientifically meaningless because of the approach used. He does not continue to rely on his specific hypotheses, but rather develops multiple fish metrics (i.e., 13 total), many of which are unrelated to his original hypothesis. My main criticisms of Stevenson's evaluation of the health of the fish community fall into the following categories:

- Lack of consideration of stream habitat characteristics in his evaluation

- Incorrect estimation of poultry house density

- Flawed statistical analysis of the data

- Flawed interpretation of the fish results

- Disregard for other available fish data for the Illinois River system.

Each of these key problems in his evaluation of the fish community of the Illinois River and its tributaries is discussed further below.

6.5.1   Lack of Consideration of Stream Habitat Characteristics in His Evaluation

A basic component of any fish survey is an evaluation of the habitat quality of the stream within the reach that is sampled. The importance of this is thoroughly discussed in Barbour et al. (1999). Stevenson (2008) makes no mention of such an evaluation. This is a surprising oversight because a habitat evaluation was to have been completed based on the fish SOP for his project, and also because Stevenson was one of the contributing authors to Barbour et al. (1999).

The habitat characteristics of sample stations were not included as part of Stevenson's analysis, which is a gross oversight. The habitat within a reach where fish sampling occurs should be evaluated, because habitat quality can greatly affect the fish community within the stream. Without adequate characterization of fish habitat it is impossible to determine if changes in the fish community at a sample station are related to habitat characteristics or other stresses related to surface water quality. Surface water quality can be ideal at a sample station, but if fish habitat is poor, the characteristics of the fish community can be dramatically affected.

Recent research in Arkansas (Dekar and Magoulick 2007) on headwater streams that are susceptible to drying has shown the importance of the stream habitat characteristics, such as size and depth of pools, in determining the structure of the fish community. In eastern Oklahoma there has been recent research by Dauwalter et al. (2007) that looks at the importance of stream morphology on the fish community present within a stream using smallmouth bass as a case example. Based on their research, the age-1 and older smallmouth bass densities were primarily determined by stream size and channel unit size. None of these important factors related to stream habitat conditions were considered by Stevenson in his analysis of the fish data.

6.5.2   Incorrect Estimation of Poultry House Density

Stevenson relies on an estimate of poultry house density that is flawed in its concept. The presence of active poultry houses is not necessarily indicative of litter application, especially for specific sub-basins in the IRW. The assumption that poultry house density can act as a surrogate for litter application rates is not supported by Stevenson (2008). In addition, Fisher's (2008) estimates of the location of active houses may not be correct, creating inaccurate poultry house density calculations. Most importantly, the poultry house density used by Stevenson (2008) was based on poultry houses located not only in the watershed, but also outside the watershed. Stevenson (2008) states that:

> In addition, poultry house density (houses/mi^2) was determined for each watershed. Observations of poultry waste application indicated most litter from poultry houses was put on fields close to its source (Fisher, personal communication). Thus, poultry houses outside the watershed of a stream could be contributing to P loading. That hypothesis was tested by relating P

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concentration in a stream to the density of poultry houses in the watershed when poultry houses within the watershed only were counted and poultry houses within 2 miles of the watershed were counted as being in the watershed. The correlation with stream P was greater when poultry houses within 2 miles of the watershed were included in the number of poultry houses in the watershed.

This is an example of Stevenson's flawed logic, which he uses to develop a construct that is then used as a primary factor in evaluating the health of the fish community. Stevenson arbitrarily assumes that counting poultry houses inside or outside of the stream watershed within a 2-mi radius of the station makes more sense because it is more closely related to phosphorus concentrations in the stream. In fact, the only poultry houses or poultry litter application that can affect a sample station are located in the watershed. Poultry houses or applications of poultry litter outside the watershed would not affect the sample station. In addition, it is only the poultry houses or litter application located upstream of a sample station within a sub-basin that can be expected to have a potential effect on the sample station. Based on Stevenson's approach, he may have counted poultry houses in areas that are located downstream of the sample station. Based on the documentation provided and my own calculations, it is not clear if he applied the 2-mile buffer to estimate poultry house density or not. His report is not consistent in the discussion of the use of poultry house density. I was able to find estimates of poultry house density only in his computer files. No documentation of how poultry house density was actually calculated was found among his files or the files of Plaintiffs' experts.

Another mistake that Stevenson (2008) made in his poultry house density estimates is that he used total poultry house numbers to estimate the densities. In his estimate, total poultry numbers included both active and inactive poultry houses. In addition, categories labeled abandoned, removed, and unknown were included in the total number, although there was ample information in Engel (2008) about the number of active poultry houses within the Illinois River and its tributaries. By counting all poultry houses to perform his density estimates, Dr. Stevenson is inflating his estimates of poultry house density. For example, the maximum poultry house density used by Stevenson (2008) was located at station RS-399 (8.2 poultry houses per square mile). At this location I evaluated the number of active poultry houses located upstream within the sub-basin of station RS-399. Using Engel's (2008) data, I calculated an active poultry house density of 4.6 poultry houses per square mile for this location.

Therefore, this estimate was considerably lower than the estimate used by Stevenson (2008). However, this number is still higher than the poultry house density calculated for station RS-399 using data provided by the individual Defendants, 3.1 poultry houses per mi^2**.**

6.5.3   Flawed Statistical Analysis of the Data

As discussed in Section 6.3, there are multiple flaws in Stevenson's (2008) statistical analysis, which make the results of his fish analysis useless. Stevenson made a judgment based on earlier statistical analyses that the percent of urban land use had an effect on the fish metrics. Therefore, he reduced his data set from 35 investigative samples down to 22 investigative samples, which biases the data to sample only locations that had 10 percent or less urban land use in the area of the site.

I independently calculated estimates of percent urban land use and my estimates matched closely with Stevenson's values. I used these estimates of urban land use with the fish metrics, sub-basin sizes, and our estimates of poultry house density at each station to determine if there was a relationship with percent urban land use, or inter-relationships between factors. As discussed in Section 5.0, there was no statistically significant relationship (or inter-relationship with other factors) between urban land use and any of the fish metrics I evaluated. Therefore, Dr. Stevenson needlessly eliminated data from his data set in a non-random way that appears to have incorporated bias in his data set. Based on Dr. Stevenson's percent urban land use criterion of 10 percent, and on review of his statistical files, it appears that he excluded the three stations (RS-654, RS-433A, and RS-757) on the main stem of the Illinois River. Thus the data set is biased, representing sampling areas off the main stem of the Illinois River with smaller sub-basins. The three sample stations on the main stem of the Illinois River had by far the largest sub-basins of the stations sampled, and as pointed out by my statistical analysis, this is the main variable affecting the fish communities within the IRW.

Dr. Stevenson relies on a statistical model he developed using poultry house density and watershed size to predict the variation of multiple fish metrics (see Table 4.1 in Stevenson [2008]). While his original hypotheses were focused on just two fish metrics related to

intolerant fish species and number of taxa, Stevenson calculated thirteen fish metrics, many unrelated to his original hypothesis. As mentioned in Section 6.3, with increasing numbers of comparisons, there is the likelihood that some comparisons will be significant by chance. For this reason it is best to limit *a priori* comparisons to those factors that are assumed to be biologically meaningful, rather than using a "shotgun" approach that simply looks at all possible combinations of dependent and independent variables. Stevenson used an unconventional (and not conservative) level of significance (P-value = 0.1) to gauge the level of significance of his results. Normally a P-value of 0.05 is used in the scientific community. Using Stevenson's 0.1 P-value, and his limited data set, only 4 of 13 metrics were significantly related to poultry house density. If the more conventional P-value of 0.05 is used, only 2 of 13 fish metrics would be significantly related to poultry house density. As mentioned in Section 6.3, if an appropriate Bonferroni correction had been applied to the level of significance to account for the 13 different metrics he evaluated, only 1 of 13 of the fish metrics would still have a significant relationship to poultry house density. At this point, most scientists would stop and not use the model's predictions if they had no statistical significance. However, Stevenson used each fish metric model (all 13 of them), even when there was no statistically meaningful relationship to poultry house density, to in fact predict the level of change in the fish metric (e.g., number of fish species) using the minimum and maximum poultry house density (see Table 4.2 of Stevenson [2008]). This is a useless statistical exercise that takes the extremes of the data set, where statistical models have the least predictive power, and tries to predict an estimate of the fish metric. As shown in Stevenson's figures (see Figures 3.1–3.3 in Stevenson [2008] as an example, based on BMI) the uncertainty in the estimates at the minimum and maximum measured value is much greater than near the middle of the measured values. Stevenson did not provide confidence intervals for any of the fish metrics in his report, which was an egregious oversight. Stevenson then averaged the maximum percent change in each fish metric to calculate an overall maximum percent change in all 13 fish metrics and called it an "average change." Based on this, he indicates that there is a 20 percent change in fish community composition. Such a calculation is meaningless as it is based on statistically insignificant relationships, so there can be no confidence that there is any relationship between poultry house density and the fish metric to begin with.

Based on my analysis of the full fish data set within the Illinois River and its tributaries, there was no statistically significant relationship between poultry house density and any of the fish metrics I evaluated. The only relationships that were determined to be statistically significant were between sub-basin size and fish community metrics (see Table 5-9). Sub-basin size was factored into Dr. Stevenson's models, but not reported in Table 4.1 in his report (Stevenson 2008). What can be concluded from the data is that sub-basin size is the primary factor affecting the fish community at a sample station in 2007.

If Dr. Stevenson's approach is used, but applied correctly to just those fish metrics that have a statistically significant relationship to sampling station sub-basin size, a percent difference in each fish metric for stream stations with a minimum and maximum sub-basin size could be calculated. The minimum and maximum sub-basin sizes for stations sampled in 2007 were selected for our estimates because this is analogous to the range used by Dr. Stevenson in his analysis. As shown in Table 5-9, percent differences in a fish metric related to sub-basin size alone range from 26 to 83 percent. These percent changes average 51 percent for five fish metrics where there is a statistically significant relationship with sub-basin size. The value of 51 percent is well above 20 percent and illustrates that a natural variable, such as sub-basin size, can have a much more powerful influence on the fish community than the 20 percent Dr. Stevenson claims is occurring as a result of changes in poultry house density. This 20 percent change is discussed further in the following section.

6.5.4   Flawed Interpretation of the Fish Results

Dr. Stevenson incorrectly used the percent change he calculated to make a flawed conclusion about the percent injury to the fish community. Stevenson (2008) states:

> Although the magnitude of species composition changes with nutrient pollution varied among indicators, the average change in indicators was approximately 20 percent. Twenty percent is an often recognized threshold for ecologically significant effects (Suter et al. 2000). Use of the 25 percent effect size as a threshold is also recommended for effluent toxicity testing (e.g., Klemm et al. 1994)

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Further, he states in his discussion that:

> These results show that poultry house activities and contaminants associated with their operations are injuring fish species composition. The 20 percent loss of most attributes across the range of poultry house densities is significant.

This is a flawed interpretation of his results even if the 20 percent change he calculated were real, which it is not. He is indicating that there is change in species composition, which means the mixture of the species has changed, not that there are not fish present. The fish data collected for the Illinois River and its tributaries fully support that there is a wide diversity of fish species in the IRW and the abundance between stations was not affected based on our statistical analysis. In addition, the use of the 20 percent as a level of injury is not accurate for measures of fish community composition. As was illustrated for a number of fish metrics, the change in a given metric within the watershed can naturally be as much as 83 percent simply based on variation in sub-basin size at the stations sampled within the IRW, regardless of the presence or absence of poultry operations. The values of 20 or 25 percent cited by Stevenson (2008) as levels of significant change are arbitrary and usually used to address the change in a specific toxic response (e.g., growth) and a specific chemical to a specific species of fish or other organism. In these latter cases, variability is reduced because of the specificity of the testing on a single organism. When evaluating the relative percent difference in fish community metrics like species diversity or abundance (represented by multiple species), the natural variability in the system can be in the range of 100 percent, as demonstrated in Table 5-9. Therefore, Stevenson's claims of injury to the fish community of the IRW are unfounded.

6.5.5   Disregard for Other Fish Data Available for the IRW

Because Stevenson (2008) based his theoretical statistical analysis on one collection of fish within 2007, it would have been appropriate to seek out other sources of information to corroborate his prediction of the health of the fish community within the Illinois River and its tributaries to see if they made sense. In this way, the results of his analysis could be compared with other investigators' results. Stevenson did not include an evaluation of other Illinois River

system fish data in his analysis. He even completely disregarded the 2005 fish data collected in the Illinois River system as part of the report, dismissing them as not fully randomized.

My evaluation considered three other sources of fish data specific to the Illinois River and its tributaries:

- The 2005 fish data collected by the Plaintiffs' consultants

- Oklahoma BUMP report information related to fish community health within the Oklahoma portion of the IRW

- EPA Region 6 specific study of the health of the fish community within the Arkansas portion of the IRW (U.S. EPA 2004).

Each of these sources of information showed that the Illinois River and its tributaries are capable of maintaining a healthy fish community in the majority of the watershed. While there are stations that do not support a cool water aquatic community or where impairment of the fish community has been found, the causes appear to be related to factors other than poultry house density (i.e., habitat alterations). These studies were more fully evaluated in Section 5.3 of my expert report. Had Stevenson (2008) considered the other data available to assess fish community health within the Illinois River system, it would have put into perspective the lack of continuity between his theoretical predications based on data collected at one point in time and the wider body of knowledge available for the Illinois River and its tributaries collected over a number of years.

6.5.6   Summary

Stevenson (2008) uses faulty statistical approaches to estimate that fish in the Illinois River system are being injured in relation to poultry house density. Taking into account the available fish data collected within the Illinois River and its tributaries, and performing my own statistical analysis of the fish data, there are no effects on the fish communities of the IRW related to poultry operations. In fact, the fish community appears healthy overall when looking more

broadly at the fish data for the Illinois River and its tributaries available from state and federal sources. My main criticisms of Stevenson's (2008) evaluation of the health of the fish community, which were discussed in detail in this section, fall into the following categories:

- Lack of consideration of stream habitat characteristics in his evaluation

- Incorrect estimation of poultry house density

- Flawed statistical analysis of the data

- Flawed interpretation of the fish results

- Disregard for other available fish data for the Illinois River and its tributaries.

While Stevenson (2008) had hypothesized that poultry house density would have an effect on the fish community within the Illinois River system, his own analysis found very few statistically significant relationships between measures of the fish community health (i.e., fish metrics) and poultry house density. The few relationships he thought he found were based on flawed statistical analysis of the data and lack of control of important variables such as sub-basin size and habitat conditions. He also overestimated poultry house density at many stations, as he counted poultry houses that were sometimes downstream of the station or not active. In addition, Stevenson (2008) removed a large portion of the available fish data before performing his final statistical analysis by arbitrarily removing any stations that were associated with greater than 10 percent urban land use. My own independent statistical analysis of the fish data showed that there was no statistically significant relationship between fish metrics and urban land use. For this reason, there was no rational reason to remove fish data from the data set. Based on my analysis, there was no statistically significant relationship between poultry house density and any of the fish metrics evaluated. My analysis of the fish data did show a statistically significant relationship between sub-basin size and many of the fish community metrics. The variation in the fish metrics from the stations with the minimum to maximum sub-basin sizes sampled within the Illinois River system varied on average by 51 percent, with the greatest percent change of 83 percent.

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Dr. Stevenson inappropriately used statistical models to predict a maximum average change of 20 percent in fish metrics. He purports that this equates to an injury to the fish community. The statistical models he relied upon were based on only a portion of the available data, and results were generally not statistically significant, so there is no confidence that the relationships upon which he based his estimates are real. In addition, as demonstrated by the effects of sampling station sub-basin size on the fish community metrics (i.e., average change of 55 percent) within the Illinois River and its tributaries, a 20 percent change in a fish metric is very small and can be accounted for by a natural variable such as sub-basin size.

Dr. Stevenson ignored other fish data collected by the State and by EPA to evaluate the health of the fish community within the Illinois River system. If he had considered these data, he would have determined that the Illinois River and its tributaries supports a vibrant warm water fish community in most areas. Based on the data presented by the state and EPA, it appears that other environmental factors, rather than poultry related operations, were the cause of stream conditions not being fully supportive of a cool water aquatic community in some parts of the IRW. These causes were related to water quality (i.e., turbidity) or likely influences from urbanization and or habitat degradation.

Taking into account the available fish data collected within the Illinois River and its tributaries, there are no apparent effects on the fish community related to poultry operations. The fish community within the Illinois River system as a whole is healthy and most areas fully support a cool water aquatic community, which includes a diverse population of game fish.

6.6   Overall Summary of Stevenson (2008)

With regard to alleged injuries to biological resources in the stream of the IRW, Stevenson (2008) used three particular aquatic communities (benthic algae, BMI, and fish) within the streams to evaluate the potential effects of nutrient loading from poultry operations on the streams. He focuses on linking increased phosphorous in surface waters of streams of the IRW with poultry operations by using an estimate of poultry house density at each station sampled within the IRW. My evaluation focused on BMI and fish communities because these

assemblages comprise widely-accepted and relevant indicators of ecosystem viability and health. Additionally, fish and BMI metrics have regulatory significance in the state of Oklahoma. Evaluations of these communities would capture any meaningful effect of changes on BMI and fish community structure caused by changes in benthic algae if such changes were present in the IRW.

I evaluated the potential for effects of poultry house density on both BMI and fish communities and found no linkage between poultry house density and the health of the BMI or fish communities in the IRW. In fact, BMI and fish communities within the IRW were healthy and diverse. Changes in BMI and fish communities found between stations or between years appeared to be related to other environmental factors rather than poultry house density. The main factors that affected BMI community composition within the IRW were sub-basin size, urban land use practices, and seasonal differences; those affecting fish community composition were sub-basin size and water quality parameters unrelated to poultry operations.

The criticisms of Dr. Stevenson's (2008) analysis have been discussed in detail in this section of my report. These problems lead to the incorrect conclusion that there was a relationship between poultry operations and the BMI and fish community composition, when in fact there was no relationship. Dr. Stevenson used his statistically flawed estimates of change in fish community metrics (i.e., 20 percent) to indicate that injury had occurred to the fish community within the IRW. These estimates (10 of 12) were calculated from regression models that included poultry house density when it was not statistically significant (i.e., not related to the fish metric). In fact, based on Dr. Stevenson's own analysis as well as my own, sub-basin size accounted for more of the change in fish metrics (significant in 8 of Dr. Stevenson's 12 models) within the Illinois River system. This highlights the meaningless nature of his conclusions, as Dr. Stevenson did not discuss the changes attributable to sub-basin size. Consequently, there is no basis on which to conclude that either the fish or BMI community of the Illinois Rover and its tributaries have been impacted by poultry house density.

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